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Neuroprotective Potential of Calycosin, Naturally Occurring Isoflavones of Formononetin

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ABSTRACT:
Flavonoids are structurally varied phytochemicals found in fruits, nuts, and vegetables, and have been linked to several health advantages. Calycosin is the most significant active flavonoid compound that is widely distributed in astragalus. various evidence suggests that calycosin exhibits anti-oxidant properties which are beneficial for neuroprotection. Calycosin has been specifically alleviated by modulating several neuroprotective pathways. In addition to this, calycosin modulates HMGB1/TLR4/NF-kappa B and NLRP3/NF-kappa B. Moreover, research indicates that calycosin promotes neuroprotection by the TRPC6/ P-CREB pathway. Besides, this calycosin also modulates neuronal apoptosis by inhibiting apoptotic protein and has another modulating action on oxidative stress and RASD1. Furthermore, several studies have reported anti-inflammatory and anti-oxidant properties of calycosin concerning neurodegenerative disease. The current review aims to provide a detailed analysis of calycosin on its mechanistic neuroprotective action.

Introduction

The field of neurodegenerative diseases presents unique challenges due to the fact that many distinctions are subtle, there are multiple clinical assessment domains (cognition, behaviour, and motor are the three main domains), and the rate at which clinical features evolve varies amongst individuals and syndromes. One of the hallmarks of neurodegenerative illnesses is the gradual accumulation of certain proteins in different brain regions [1]. The incidence and prevalence of AD that has been clinically identified at different stages point to the areas where health planners and physicians might intervene. According to evidences, compared to normal persons (84% of participants: 3,926 cognitively unimpaired), the subjects exhibiting clinical symptoms comprise a very small number (16% of participants: 640 MCI and 94 dementia).In a restricted sample of around 300 patients, research conducted in Goteborg, Sweden found that the prevalence of amyloid pathology was 22.8%, t-tau pathology was 33.2%, and p-tau pathology was 6.9% among those with a CDR score of 0 (normal persons) [2].

Since 1990, there has been a 39% rise in neurological disorder-related mortality (34–44) and a 15% increase in disability-adjusted life years (DALYs) (9–21). Nonetheless, over the same period, age-standardized death rates dropped by 28% (26–30).Between 1990 and 2016, there was a 27% (24–31) decline in the age-

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standardized DALY rates [3]. From 1990 to 2019, there were rising trends in the prevalence of Parkinson's disease (PD) in most areas and nations worldwide.[4]. Various evidences indicates that cytokines play a role in modulating responses to peripheral nerve injury, controlling behaviour, mediating physiological sleep and synaptic plasticity, advancing or inhibiting neurodegeneration, and communicating systemic injury, infection, and inflammation to the brain [5].

The hydroxy isoflavone compound calycosin, also known as 7-hydroxy-3-(3-hydroxy-4-methoxyphenyl) chrome-4-one, is frequently isolated from the dry root extract of Radix astragali [6]. However, several studies have reported that calycosin has the ability to inhibit GSK-3β activity, which in turn reverses the hyperphosphorylation of tau and inhibits the production of RAGE and BACE-1 proteins [7]. On the other hand, a number of studies have shown that calycosin plays a critical role in pS6K and p4EBP1 levels, indicating that anabolic and catabolic processes are kept in equilibrium and that this prevents neuronal cell death [8]. However, several studies have documented that calycosin lowers MDA, ROS, SOD, and GSH-Px levels, which has a neuroprotective impact against cerebral ischemia/reperfusion damage to confer neuroprotection [9]. Therefore, by supporting published reports the neuroprotecting capabilities of calycosin in this review aim to give mechanistic neuroprotective potential.

Calycosin promotes neuroprotection by TRPC6/ P-CREB pathway

TRPC6 is a Ca^{2+} permeable non-selective cation channel that is expressed in our brain [10]. TRPC6 mRNA level in the blood cells were found to decrease, leading to cognitive dysfunction [11]. TRPC6 promotes the phosphorylation of Camp/Ca^{2+} and phosphorylation of CREB, further downregulation of TRPC6 mRNA expression which plays a crucial role in the pathology of neurodegeneration[12]. TRPC6 increased the Ca^{2+} concentration which may activate these CREB in 3 signaling cascade 1.) Ras/MEK/ERK, 2.) RAS/PI3K/AKT & 3.) CaM/CAMKIV. These signaling cascades may activate the MAPK which leads to CREB phosphorylation further it regulates the BDNF expression that possesses neuroprotection [13]. Several animal model studies have reported the role of TRPC6 in neuroprotection via CREB phosphorylation. By using MCAO-induced brain injury Guo.et.al documented that calycosin (20mg/kg) significantly improved the decreased neurological deficits. By using histopathological analysis calycosin (20 mg/kg) decreased the increased water content and necrotic neurons in the brain. Furthermore, By using western blot analysis, calycosin (20 mg/kg) significantly decreased the increased positive cells in the penumbral cortex. It also reversed the reduced TRPC6 & P-CREB and significantly weakened the calpain activity. The author also demonstrated that the administration of SKF96365 abolished the improvement of neurological deficits and significantly increased the infarct area in the brain of SKF96365 rats. By using CCK-8 and flow cytometry analysis author documented that it alleviated the neuronal cell death and cell apoptosis induced by OGD injury. It also reduced the increased Ca^{2+} concentration induced by OGD injury. By using western blot analysis author documented that calycosin prevents the decrease of TRPC 6 & P-CREB levels in the brain of rats [14]. Overall evidence suggests that calycosin might promote neuroprotection by TRPC6/ P-CREB pathway.

Modulation of neuroinflammation by HMGB1/TLR4/NF-kappa B and NLRP3/NF-kappa B

HMGB1 is a highly conserved ubiquitous protein [15]. Evidence also delineates that HMGB1 is released from intracellular space to extracellular space. It interacts with TLR4 directly and with RAGE on the surface of neurons. HMGB1/ TLR4 signaling pathway provides its signal through the My-88 pathway. HMGB1 binds to TLR4, a member of a dangerous associated molecular pattern family, and provides a danger signal that further activates the innate immune system and possesses an inflammatory response [16]. Activation of HMGB1 triggers the activation of NF-κB and activator protein (AP1), via the MyD-88 pathway which promotes the induction of NLRP3 inflammasome and cytokines like as IL-1B, IL-6, IL-8, IL-12, TNF-α which leads to neurodegeneration[17]. By using OGD rat Li.et.al documented that calycosin (4μm) significantly increased the decreased cell viability of microglial cells in BV2 & HAP1 cells. By using ELISA author demonstrated that calycosin (4μm) reduced the increased IL-1b, IL-6 & TNF-a in OGD rats. By using ELISA, western blot analysis, and QT/PCR calycosin reduced the increased

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TLR4 protein level in OGD rats. It also inhibits the phosphorylation level of NF-kB & IkBa in ODG rats. By using BV2 cells author also documented calycosin reduced the increased mRNA levels of HMGB1 & TLR4 of ODG rats [18].By using MPTP-induced Parkinsons's disease Yang. et.al documented that calycosin (15 mg/kg) increased the reduced TH immunoreactive and inhibited the necrosis of neurons. It also reduced the increased CD11b levels. By using RT/PCR author documented that it reduced the mRNA expression level of TNF-1b, IL-1b, and IL-6 in Parkinson's disease mice. Using LPS-induced BV-2 cells it reduced the increased phosphorylation level of P38, JNK, & ERK. By using western blot analysis it reduced the increased TLR2, TLR4 & NF-kb in LPS-induced BV2 cells. By using LPS-induced BV-2 cells author documented that it reduced the mRNA expression level of TNF-1b, IL-1b, and IL-6 in Parkinson's disease mice. By using immunohistochemistry analyses author documented that calycosin attenuated the increased Tnf-a level [19]. By using ICH induced oxidative stress mice model Chen.et.al documented that calycosin (10, 15, 50, 75 mg/kg) significantly increased the decreased the NDS score. It also repressed the 4-HNE level and increased the Nrf2, Sod 1 in calycosin-treated mice. Furthermore, By using ELISA, also reduced the transcription and translation level of NLRP3, Caspase-1, IL-1b, & IL-18. The author also demonstrated that it significantly reduced the increased level of TNF-a, IL-6, NF-kb & P65 [20].By using immunohistochemistry analyses author documented that calycosin attenuated the increased Tnf-a level [21]. Evidence suggests that once activated the HMGB1/TLR/NF-kB and NLRP3/NF-kb produce an inflammatory response and pro-inflammatory cytokines. Overall, evidence suggests that calycosin might reduce neuroinflammation by inhibiting HMGB1/TLR4/NF-kB & NLRP3/NF-Kb mediated production of pro-inflammatory cytokines.

Modulation of neuronal apoptosis by inhibiting apoptotic protein

Apoptosis is also known as cell death. Apoptosis is based on its proteins which come under the Bcl-2 family and this family includes both inhibitors and modifiers of apoptosis. Inhibitors of Apoptosis are Bcl-2, Bcl-X, and Mcl-1 and modifiers are BAX, BCL-X_5, BAD, BAK, and BIK. Several evidence delineate that the mRNA level of Bcl-2 protein is decreased which promotes neurodegeneration [22]. The bcl-2/Bax ratio determines the apoptosis of cell, upregulation of BAX is accompanied by the downregulation of the Bcl-2 protein to promote caspase-3 pathway which possesses apoptosis [23]. Bcl-2 family plays a crucial role in promoting or inhibiting intrinsic apoptosis which is trigerred by dysfunction of mitochondria [24]. SIRT 1 an NAD+ dependent protein while PG-a is a nuclear transcriptional co-activator [25].Increased SIRT1 inhibit apoptosis by regulating p53 & by deacetylating PGC-a [26]. By utilizing different animal model several studies have deleniated the neuroprotective potential of calycosin through modulation of neuronal apoptosis. By using the MCAO model Wang.et.al demonstrated that calycosin (20mg/kg) reduced the neurological deficits score and also ameliorated the brain dysfunction. Furthermore, it also reduced the infarct area and brain water content. By using RT/PCR and western blot analysis author documented that calycosin increased the Bcl-2 and mRNA levels in the cerebral part of the brain [21]. It also reduced the apoptosis caused by OGD/R. Furthermore, by using western blot analysis calycosin increased the reduced SIRT 1, FOXO1, PGC-α and Bcl-2 protein level. It also reduced the Bax expression level [26]. By using MCAO-inducing injury wang .et.al documented that calycosin (20mg/kg) reduced the increased infarct volume and brain edema in the brain. It also increased the decreased expression of p-P13K/PI3K, p-AKT/AKT, and Bcl-2/BAX, but also reduced the expression of GSK-3b in the ischemic brain. It also enhanced the Bcl-2 , ER-α and miR-375 mRNA expression [27]. Overall, evidence suggests that calycosin might reduce neuroinflammation by inhibiting apoptotic protein . By using MCAO-induced cerebral ischemia injury fu.et.al demonstrated that calycosin (25 mg/kg) decreased the increased infarct size and tissue damage in the brain of rats. It also reduced the BBB permeability. By using western blot analysis author demonstrated that calycosin increased the reduced ZO-1 and claudin-5 in the ischemic brain tissue of ischemia-reperfused brains. Furthermore, the author also documented that it significantly reduced MMPs activities and also down-regulated the expression of MMP-2 and MMP-9. It also increased the decreased expression level of cav-1 in the brain. Calycosin also inhibited the intensity of NO in the DETA/NO treated bEnd cells [28].

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Modulation of oxidative stress by inhibiting ROS/RNS and RASD1

Oxidative stress is common cause for neurodegeneration which results from the unregulated production of ROS and these ROS are produced by mitochondria. These mitochondrial ROS may possesses apoptosis and may also impair mitochondrial dynamics via autophagy [29]. Much evidence shows that oxidative stress plays a critical role in neurodegeneration by producing reactive nitrogen species, further, these ROS and RNS decreased Antioxidant enzymes such as SOD, and GPx in models of neurodegeneration [30,31]. RASD1 is a member of the RAS superfamily of small G proteins [32]. RASD1 inhibits the Camp-PKA-CREB pathway after inhibition of this pathway the phosphorylation of CREB is inhibited, production of BDNF reduced by which neurodegeneration occurs [33]. By utilizing different animal model several studies have deleniated the neuroprotective potential of calycosin by inhibiting oxidative stress. By using MCAO-induced injury guo.et.al documented that calycosin (15 mg/kg) decreased the increased neurological deficits score. It also reduced the increased infarct volume in MCAO injury mice. Moreover, it also decreased the increased ROS and carbonyl protein content in rats. Furthermore, it also increased the decreased activities of SOD, catalase, and GSH-Px and decreased the MDA content in the brain part of the brain. By using immunohistochemical staining calycosin significantly decreased the increased immunopositive cells [34]. By using OGD-induced injury the yan.et.al documented that calycosin (10 mg/kg) alleviated the increased cell death and LDH leakage in OGD-induced rats. Moreover, it also significantly reverted the increased level of MDA and ROS and decreased SOD activity in the hippocampal region of the brain. Furthermore, It also reverted the decreased cell viability, SOD, and increased level of ROS, LDH, & MDA [26]. It also significantly decreased the increased apoptosis of the cell [35].

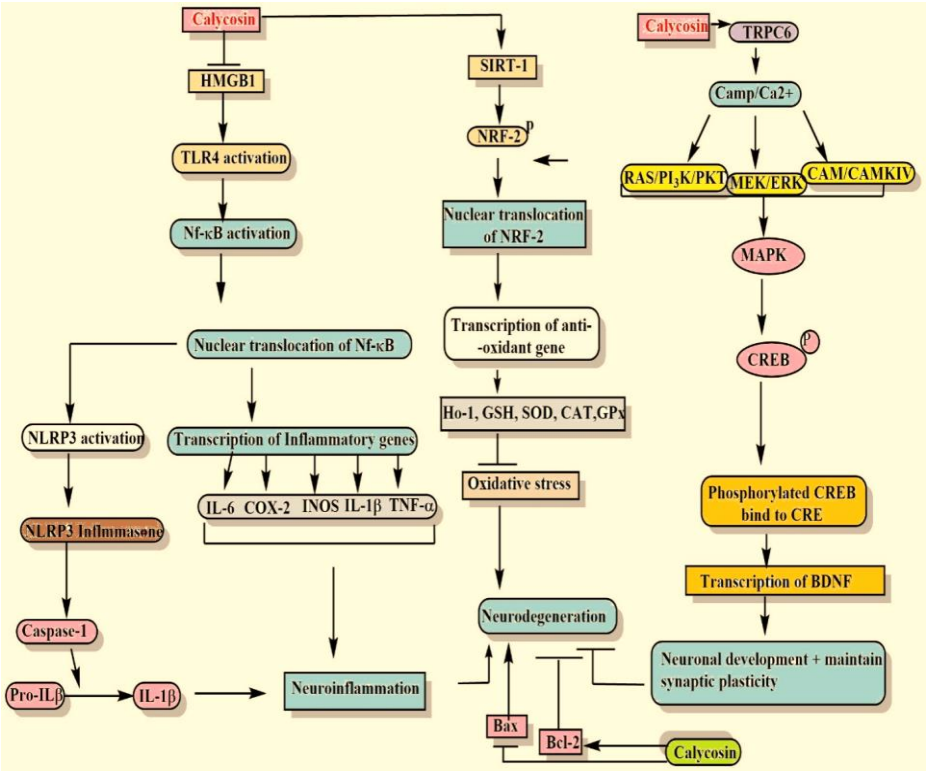

Figure no:-1 Calycosin modulates various mediators like HMGB1, TLR4 , SIRT, TRPC, oxidant , antioxidant and various inflammatory mediators to confor neuroprotection. Calycosin inhibit the HMGB1/TLR4 activation thus resulting in downregulation of NF-κB mediated pro-infammatory cytokines such as IL-6, Cox-2, Inos, TNF-α. Calycosin potentiate the SIRT1 that activate the NRF-2 phosphorylation leads to

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transcription of anti-oxidant gene such as SOD, GSH, CAT, GPx, these antioxidant suppress the oxidative stress and help in neuroprotection. Calycosin activate the TRPC6 protein which activate the Camp/Ca^{2+} this pathway activate the MAP kinase via PI3K/PKT, MEK/ERK & CAM/CAMKIV that's leads to phosphorylation of CREB and bind with CRE for transcription of BDNF that potentiate neuroprotection. Calycosin inhibit Bax and activate Bcl-2 to potentiate neuroprotection.

By using the APP/PS1 mice model song.et.al documented that calycosin(40mg/kg) significantly increased the time spent and escape latency time and retention time in APP/PS1 mice. Moreover, it also decreased the increased amyloid beta, tau protein, TNF-α, IL-1β, AChE, and MDA levels but it increased the decreased GSH level in APP/PS1 mice [36]. By using cerebral ischemia/ reperfusion rats wang .et.al documented that calycosin (5 mg/kg) decreased the increased infarct volume and RASD1 mRNA expression [35]. By using l-glutamate-induced toxicity yu.et.al documented that calycosin decreased the increased radicals and increased the decreased GSH, SOD, and GSH-Px levels in PC-12 cells [37]. Overall, evidence suggests that calycosin might reduce neuroinflammation by inhibiting oxidative stress and RASD1.

Table no 1: Summary of the neuroprotective effect of calycosin in various in vivo models.

SR. N0	ANIMAL MODEL	DOSE	OBSERVED EFFECT	REFERENCE
1.	MCAO-Induced brain injury mice mo	20mg/kg	Decrease the positive cells in the cerebral cortex and water content Reduce TRPC6 & CREB. Decrease neuronal death.	[14]
2.	ODG rat model	4um	Decrease IL-1b, IL-6 & TNF-a Decrease the TLR4 protein Inhibit phosphorylation of NF-kB	[18]
3.	MPTP induced parkinsons mice model	15 mg/kg	Decrease the CD11b level Decrease IL-1b, IL-6 & TNF-a	[19]
4.	ICH-induced oxidative stress mice model	10,25,50&75mg/kg	Decrease the neurological deficits score Reduce TNF-a, IL-6, NF-kb & P65 InhibiHMGB1/TLR4/NF-kB & NLRP3/NF-kB	[20]
5.	OGD/R induces oxidative stress and neuronal apoptosis mice model	20mg/kg	Inhibit infarct area and water content Increase SIRT 1, FOXO1, PGC-a and Bcl-2 protein Increase p-P13K/PI3K, P-AKT & Bcl-2/BAX.	[26]
6.	MCAO/R Induce ischemic injury and reperfusion rat model	20 mg/kg	Increase p-P13K/PI3K, P-AKT & Bcl-2/BAX. Down regulate the level ROS, LDH, MDA.	[27]
7.	MCAO-induced Cerebral ischemia rat model	15mg/kg	Reduce neurological deficits score Increase SOD, catalase, GSH, Gpx.	[34]
8.	Ischemia and reperfusion induced neurological impairment rat model	10mg/kg	Decrease the Bcl- Rna level. Decrease the TNF-a level.	[21]

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9.	APP/PS1 mice model	40mg/kg	Reduce the amyloid beta, tau protein, TNF-a, IL-1B, AChE, and MDA levels Increase the GSH level	[36]
10.	Cerebral ischemia/ reperfusion rat model	5mg/kg	Decreased the infarct volume Decreased the RASD1 level Increase GSH,SOD &GSH-PX	[35]
11.	L-glutamate-induced toxicity rat model	(0.05g/ml)	Increase GSH, SOD, GSH-Px level	[37]

Conclusion

In summary, this review aims to highlight the neuroprotective potential of calycosin, promoting their neuronal health of their mechanistic neuroinflammatory pathway. However, calycosin activate the TRPC6 protein which activate the Camp/Ca^{2+}, leads to phosphorylation of CREB and bind with CREB for transcription of BDNF that potentiate neuroprotection. Additionally, calycosin explores the potential synergistic effects in neuroprotection for novel therapeutic interventions for neurodegenerative disease. Therefore, the future prospects involving mechanistic action of calycosin underlying the neuroprotective effects and several novel studies validate their efficacy for exploring their potential for neurodegenerative disease.

Conflict of Interest

There is no conflict of interest.

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Reference

1. Cornblath E.J., Robinson J.L., Irwin D.J., Lee E.B., Lee V.M.Y., Trojanowski J.Q., Bassett D.S., 2020. Defining and predicting transdiagnostic categories of neurodegenerative disease. Nat Biomed Eng. 4(8), 787-800.
2. Kern S., Zetterberg H., Kern J., Zettergren A., Waern M., Höglund K., Skoog I., 2018. Prevalence of preclinical Alzheimer disease: comparison of current classification systems. Neurology. 90(19), e1682-e1691.
3. GBD 2016 Neurology Collaborators 2019. Global, regional, and national burden of neurological disorders, 1990-2016: a systematic analysis for the Global Burden of Disease Study 2016. Lancet Neurol. 18(5), 459–480.
4. Ou Z., Pan J., Tang S., Duan D., Yu D., Nong H., Wang Z., 2021. Global Trends in the Incidence, Prevalence, and Years Lived With Disability of Parkinson's Disease in 204 Countries/Territories From 1990 to 2019. Front Public Health. 9, 776847.
5. Merrill J. E., Benveniste E. N., 1996. Cytokines in inflammatory brain lesions: helpful and harmful. Trends Neurosci. 19(8), 331-338.
6. Gao J., Liu Z. J., Chen T., Zhao D., 2014. Pharmaceutical properties of calycosin, the major bioactive isoflavonoid in the dry root extract of Radix astragali. Pharm Biol. 52(9), 1217-1222.
7. An F., Zhao R., Xuan X., Xuan T., Zhang G., Wei C., 2022. Calycosin ameliorates advanced glycation end product-induced neurodegenerative changes in cellular and rat models of diabetes-related Alzheimer's disease. Chem Biol Interact. 368, 110206.
8. Chaouhan H.S., Li X., Sun K.T., Wang I.K., Yu T.M., Yu S.H., Li C.Y., 2022. Calycosin alleviates paraquat-induced neurodegeneration by improving mitochondrial functions and regulating autophagy in a drosophila model of Parkinson’s disease. Antioxidants. 11(2), 222.
9. Guo C., Tong L., Xi M., Yang H., Dong H., Wen A., 2012. Neuroprotective effect of calycosin on cerebral ischemia and reperfusion injury in rats. J Ethnopharmacol. 144(3), 768-774.
10. Dietrich A., Gudermann T., 2007. TRPC6. Handb Exp Pharmacol. (179), 125-41.
11. Zernov N., Popugaeva E., 2023. Role of Neuronal TRPC6 Channels in Synapse Development, Memory Formation and Animal Behavior. Int J Mol Sci. 24(20), 15415.

Journal of Chemical Health Risks

www.jchr.org

JCHR (2024) 14(3), 862-869 | ISSN:2251-6727

12. Yao C., Zhang J., Chen F., Lin Y., 2013. Neuroprotectin D1 attenuates brain damage induced by transient middle cerebral artery occlusion in rats through TRPC6/CREB pathways. Mol Med Rep. 8(2), 543–550.
13. Chen G., Ward M.F., Sama A.E., Wang, H., 2004. Extracellular HMGB1 as a proinflammatory cytokine. J Interferon Cytokine Res. 24(6), 329–333.
14. Guo C., Ma Y., Ma S., Mu F., Deng J., Duan J., Xiong L., Yin Y., Wang Y., Xi M., Wen A., 2017. The Role of TRPC6 in the Neuroprotection of Calycosin Against Cerebral Ischemic Injury. Sci Rep. 7(1), 3039.
15. Zhang S., Chen F., Zhai F., Liang S., 2022. Role of HMGB1/TLR4 and IL-1β/IL-1R1 Signaling Pathways in Epilepsy. Front Neurol. 13, 904225.
16. Luo L., Wang S., Chen B., Zhong M., Du R., Wei C., Huang F., Kou X., Xing Y., Tong G., 2022. Inhibition of inflammatory liver injury by the HMGB1-A box through HMGB1/TLR-4/NF-κB signaling in an acute liver failure mouse model. Front Pharmacol. 13, 990087.
17. Tatton W.G., Olanow C.W., 1999. Apoptosis in neurodegenerative diseases: the role of mitochondria. Biochim Biophys Acta. 1410(2), 195-213.
18. Li X., Yang X., Lu H., Wang W., Cai L., Chen J., Wang Y., 2023. Calycosin attenuates the inflammatory damage of microglia induced by oxygen and glucose deprivationthrough the HMGB1/TLR4/NF-κB signaling pathway: calycosin attenuates OGD/R damage inmicroglia. Acta Biochim Biophys Sin. 55(9), 1415.
19. Yang J., Jia M., Zhang X., Wang P., 2019. Calycosin attenuates MPTP-induced Parkinson's disease by suppressing the activation of TLR/NF-κB and MAPK pathways. Phytother Res. 33(2), 309-318.
20. Chen C., Cui, J., Ji X., Yao L. 2020. Neuroprotective functions of calycosin against intracerebral hemorrhage-induced oxidative stress and neuroinflammation. Future Med Chem. 12(7), 583-592.
21. Wang Y., Ren Q., Zhang X., Lu H., Chen J., 2018. Neuroprotective mechanisms of calycosin against focal cerebral ischemia and reperfusion injury in rats. Cell Physiol Biochem. 45(2), 537-546.
22. Schroer J., Warm D., De Rosa F., Luhmann H. J., Sinning A., 2023. Activity-dependent regulation of the BAX/BCL-2 pathway protects cortical neurons from apoptotic death during early development. Cell Mol Life Sci. 80(6), 175.
23. Zhang Y., Yang X., Ge X., Zhang F., 2019. Puerarin attenuates neurological deficits via Bcl-2/Bax/cleaved caspase-3 and Sirt3/SOD2 apoptotic pathways in subarachnoid hemorrhage mice. Biomed Pharmacother. 109, 726-733.
24. Zhou Y., Wang S., Li Y., Yu S., Zhao Y., 2018. SIRT1/PGC-1α Signaling Promotes Mitochondrial Functional Recovery and Reduces Apoptosis after Intracerebral Hemorrhage in Rats. Front Mol Neurosci. 10, 443.
25. Singh P., Hanson P.S., Morris C.M., 2017. SIRT1 ameliorates oxidative stress induced neural cell death and is down-regulated in Parkinson's disease. BMC Neurosci. 18, 1-13.
26. Yan X., Yu A., Zheng H., Wang S., He Y., Wang L., 2019. Calycosin-7-O-β-D- glucoside attenuates OGD/R-induced damage by preventing oxidative stress and neuronal apoptosis via the SIRT1/FOXO1/PGC-1α pathway in HT22 cells. Neural Plast. 2019, 8798069.
27. Wang P. C., Wang S. X., Yan X. L., He Y. Y., Wang M. C., Zheng H. Z., Wang L.S., 2022. Combination of paeoniflorin and calycosin-7 glucoside alleviates ischaemic stroke injury via the PI3K/AKT signalling pathway. Pharm Biol 60(1), 1469-1477.
28. Fu W., Che X., Tan J., Cui S., Ma Y., Xu D., Long H., Yang X., Wen T., He Z., 2024. Rasd1 is involved in white matter injury through neuron-oligodendrocyte communication after subarachnoid hemorrhage. CNS Neurosci Ther 30(3), e14452.
29. Teleanu D.M., Niculescu A.G., Lungu I.I., Radu C.I., Vladâcenco O., Roza E., Costăchescu B., Grumezescu A.M., Teleanu R.I., 2022. An Overview of Oxidative Stress, Neuroinflammation, and Neurodegenerative Diseases. Int J Mol Sci. 23(11), 5938.
30. Fernández-Checa J.C., Fernández A., Morales A., Marí M., García-Ruiz C., Colell A., 2010).

Oxidative stress and altered mitochondrial function in neurodegenerative diseases: lessons from mouse models. CNS Neurol Disord Drug Targets. 9(4), 439–454.

31. Pong K., 2003. Oxidative stress in neurodegenerative diseases: therapeutic implications for superoxide dismutase mimetics. Expert Opin Biol Ther. 3(1), 127–139.
32. Fesharaki-Zadeh A., 2022. Oxidative Stress in Traumatic Brain Injury. Int J Mol Sci. 23(21), 13000.
33. Gao S., Jin L., Liu G., Wang P., Sun Z., Cao Y., Shi H., Liu X., Shi Q., Zhou X., Yu R., 2017. Overexpression of RASD1 inhibits glioma cell migration/invasion and inactivates the AKT/mTOR signaling pathway. Sci Rep. 7(1), 3202.
34. Guo C., Tong L., Xi M., Yang H., Dong H., Wen A., 2012. Neuroprotective effect ofcalycosin on cerebral ischemia and reperfusion injury in rats. J Ethnopharmacol. 144(3), 768-774.
35. Wang Y., Dong X., Li Z., Wang W., Tian J., Chen J. 2014. Downregulated RASD1and upregulated miR-375 are involved in protective effects of calycosin on cerebral ischemia/reperfusion rats. J Neurol Sci. 339(1-2), 144-148.
36. Song L., LiX., Bai X.X., Gao J., Wang C.Y., 2017. Calycosin improves cognitive function in a transgenic mouse model of Alzheimer's disease by activating the protein kinase C pathway. Neural Regen Res. 12(11), 1870-1876.
37. Yu D., Duan Y., Bao Y., Wei C., An L., 2005. Isoflavonoids from Astragalus mongholicus protect PC12 cells from toxicity induced by L-glutamate. J Ethnopharmacol. 98(1-2), 89-94.