

Based on Network Pharmacology to Explore the Mechanism of Action of *Achyranthes Bidentata* in Treating Stroke

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Abstract:

Purpose: To analyze the mechanism of intervention of *Achyranthes bidentata* on stroke through network pharmacology techniques. **Methods:** The active components and potential targets of *Achyranthes bidentata* were screened using the Traditional Chinese Medicine Systems Pharmacology (TCMSP) database and analysis platform. Stroke-related targets were screened in gene-related databases (GeneCards, OMIM, DrugBank, and TTD). Shared targets between *Achyranthes bidentata* and stroke were obtained through online Venny analysis, and a visualization network of the *Achyranthes bidentata*–stroke shared targets was constructed using Cytoscape. A protein-protein interaction (PPI) network was built with the STRING database, followed by topological analysis of the shared targets to identify core targets. Using RStudio, the top 10 GO terms and the top 30 KEGG pathways for the shared targets were selected and visualized for enrichment analysis. **Result:** From the database, 20 active constituents of *Achyranthes bidentata* and 186 corresponding targets were identified, with 127 of these targets overlapping with those implicated in stroke. The herb's key active compounds include quercetin, kaempferol, and wogonin. Core targets highlighted by the analysis encompass TP53, MAPK1, AKT1, among others. Gene ontology enrichment points to biological processes such as response to molecules of bacterial origin, response to lipopolysaccharide, and response to xenobiotic stimulus. KEGG pathway enrichment suggests that lipid metabolism and atherosclerosis, the AGE-RAGE signaling pathway, and fluid shear stress-related atherosclerosis may underlie the mechanisms by which *Achyranthes bidentata* intervenes in stroke. **Conclusion:** *Achyranthes bidentata* may exert therapeutic effects on stroke through multiple constituents acting on diverse targets.

Keywords:

Network Pharmacology, Stroke, Mechanism, *Achyranthes Bidentata*

1. Introduction

Stroke, a common neurological condition among middle-aged and elderly individuals, mainly presents as localized neurological deficits resulting from cerebrovascular events, with high rates of disability and death [1,2]. Its development involves multiple pathological processes, including neuroinflammation, disrupted energy metabolism, and compromised microvascular barriers. Modern medical approaches emphasize acute phase strategies such as restoring blood flow, preventing blood clots, and protecting neural functions [3]. In traditional Chinese medicine, the disease is primarily attributed to dysfunction of internal organs leading to disrupted qi and blood flow, along with pathogenic factors like wind, fire, phlegm, and blood stasis impairing mental clarity [4]. Epidemiological data indicate that phlegm and stasis-related syndromes are common among stroke patients. Research shows that Chinese herbal medicines can treat stroke through various mechanisms, including reducing oxidative stress, preventing cell death, and regulating blood vessel function. Notably, *Achyranthes bidentata* is a key herb known for its abilities to promote blood circulation, eliminate stasis, open meridians, and relieve pain [5]. This study utilizes network pharmacology to analyze *Achyranthes bidentata*'s active ingredients, molecular targets, and signaling pathways, providing a detailed understanding of how it works against stroke, and offering new insights into traditional Chinese medicine's role in cerebrovascular disease prevention and treatment.

2. Materials and Methods

2.1. Screening of active components in *Achyranthes bidentata* and prediction of related targets

The active compounds in *Achyranthes bidentata* were identified by screening the Traditional Chinese Medicine Systems Pharmacology Database (TCMSP) using specific pharmacokinetic criteria—oral bioavailability (OB) $\geq 30\%$ and drug-likeness (DL) ≥ 0.18 . The relevant protein targets associated with these bioactive components were then extracted and annotated using the UniProt database for gene identification.

2.2. *Achyranthes bidentata*—Shared Targets in Stroke and Network Construction

Using “stroke” as the keyword, search GeneCards, OMIM, DrugBank, and TTD databases, then integrate and deduplicate to obtain disease targets. Apply the online tool Venn 2.1.0 to analyze and process the targets of the Chinese herbal components and the disease targets, ultimately identifying the shared targets. Employ Cytoscape to build a network diagram illustrating the relationship between the targets of *Achyranthes bidentata* and stroke, showing the commonConstructing a Protein-Protein Interaction (PPI) Network for *Achyranthes bidentata* and Stroke

2.3. Constructing the PPI Network for *Achyranthes bidentata*—stroke-related targets

Inputting the *Achyranthes bidentata*—stroke related target genes into the STRING database, selecting *Homo sapiens* as the species, to generate a PPI network. Then, employing Cytoscape for topological analysis, we took the degree of connectivity twice to refine and identify the key core targets within the network.

2.4. *Achyranthes bidentata*—stroke shared target GO analysis and KEGG pathway enrichment analysis

Import the *Achyranthes bidentata*—stroke shared targets into RStudio for GO functional enrichment analysis and KEGG pathway enrichment analysis. The GO analysis includes Biological Process (BP), Molecular Function (MF), and Cellular Component (CC). Following a criterion of $P \leq 0.05$ and sorting from smallest to largest, select the top 10 GO analyses and the top 30 KEGG pathway enrichment results, and visualize the analysis results.

3. Result

3.1. Active components of *Achyranthes bidentata* and related targets

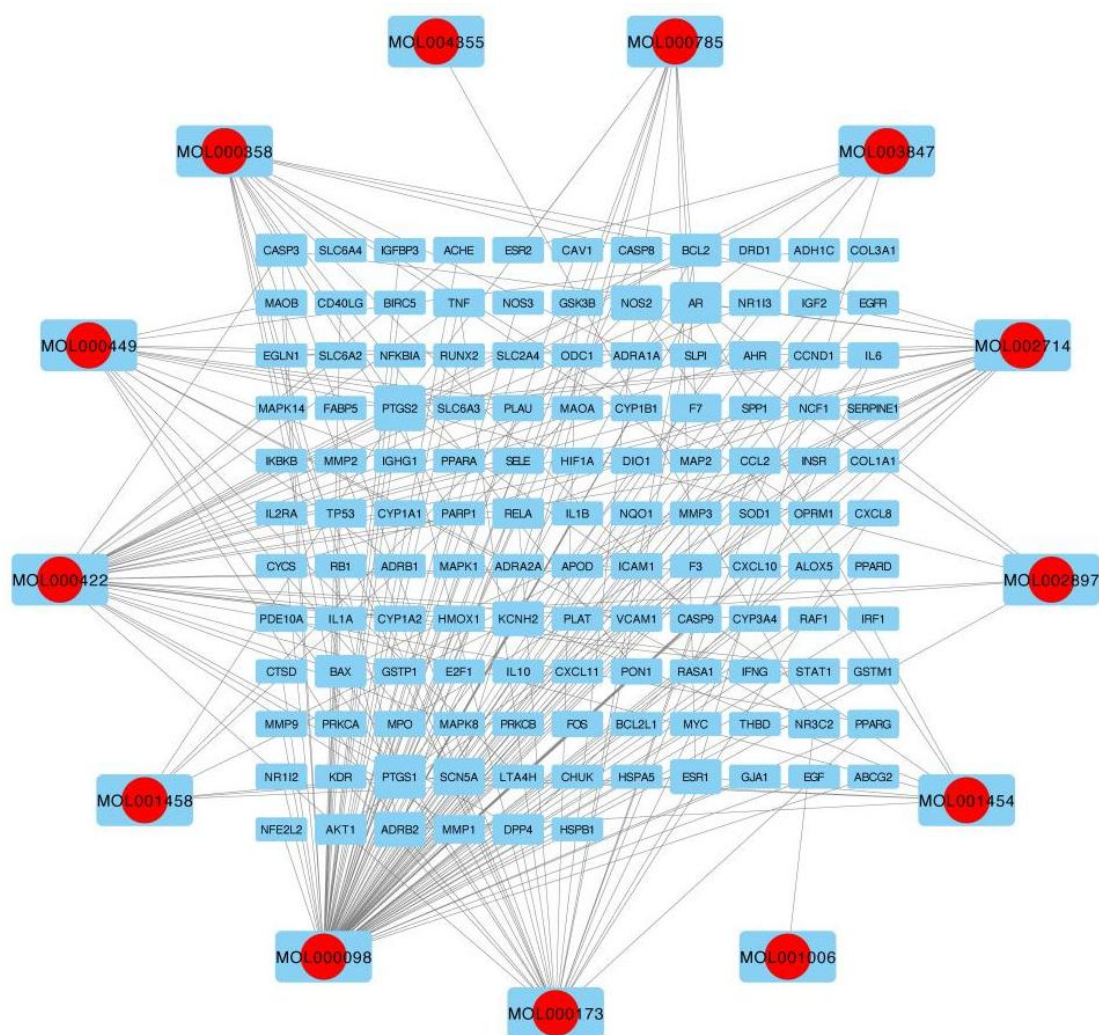


Figure 1. Illustration of Compound-Target Gene Interactions Network.

The study identified 20 bioactive compounds linked to 186 potential targets. After filtering out non-active components, 127 key targets associated with *achyranthes bidentata* were isolated. A network visualization was created (Figure 1) depicting these relationships, with bioactive compounds shown in the outer ring and their corresponding targets in the inner ring. The connections were weighted by interaction strength, revealing four primary active constituents based on their degree centrality: kaempferol (MOL000422), quercetin (MOL000098), wogonin (MOL000173), and stigmasterol (MOL000449). These compounds emerged as the core bioactive elements through network analysis.

To gain a more comprehensive understanding of *Achyranthes bidentata*'s therapeutic potential in stroke treatment, we conducted an in-depth analysis of the 127 candidate targets previously identified. This was achieved using the String database, with the results visualized in Figure 3.

Leveraging the CytoNCA plug-in, we performed a detailed topological assessment of the protein interaction network. Our analysis pinpointed critical hub targets that met stringent criteria, including BC values surpassing 7.62, CC values exceeding 0.52, DD values above 8, EC values greater than 0.15, LAC values beyond 4.18, and NC values higher than 5.43. From this, we isolated the core targets and determined that the top five most influential in *Achyranthes bidentata*'s stroke treatment were TP53, MAPK1, AKT1, TNF, and FOS, among others. These findings are further illustrated in Figure 4.

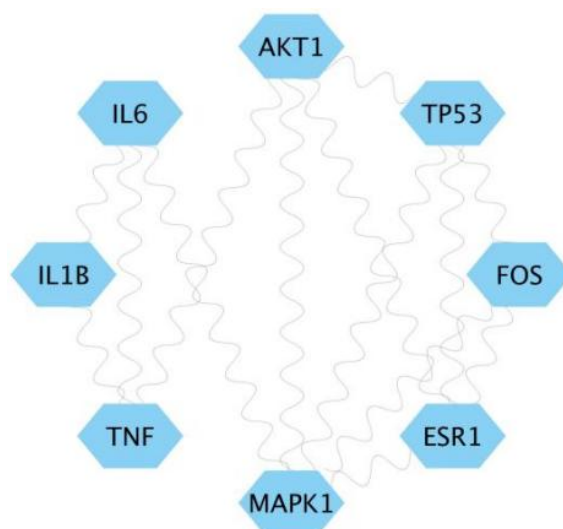


Figure 4. PPI core network screening.

3.4. Analysis Results of Go and KEGG Pathway Enrichment for *Achyranthes bidentata* in treating stroke

Using the R language software, a total of 2388 GO terms were enriched, including 2158 BP terms, 38 CC terms, and 192 MF terms. These mainly involved biological processes such as response to external stimulus, response to hypoxia, and response to oxygen level; cellular components such as membrane raft, membrane microdomain, and caveola; molecular functions such as DNA-binding transcription factor binding, RNA polymerase II-specific DNA-binding, transcription factor binding, and nuclear receptor activity (Figure 5). Additionally, a total of 188 KEGG pathways were enriched. These mainly included Lipid and atherosclerosis, AGE-RAGE signaling pathway in diabetic complications, and Fluid shear stress and atherosclerosis (Figure 6).

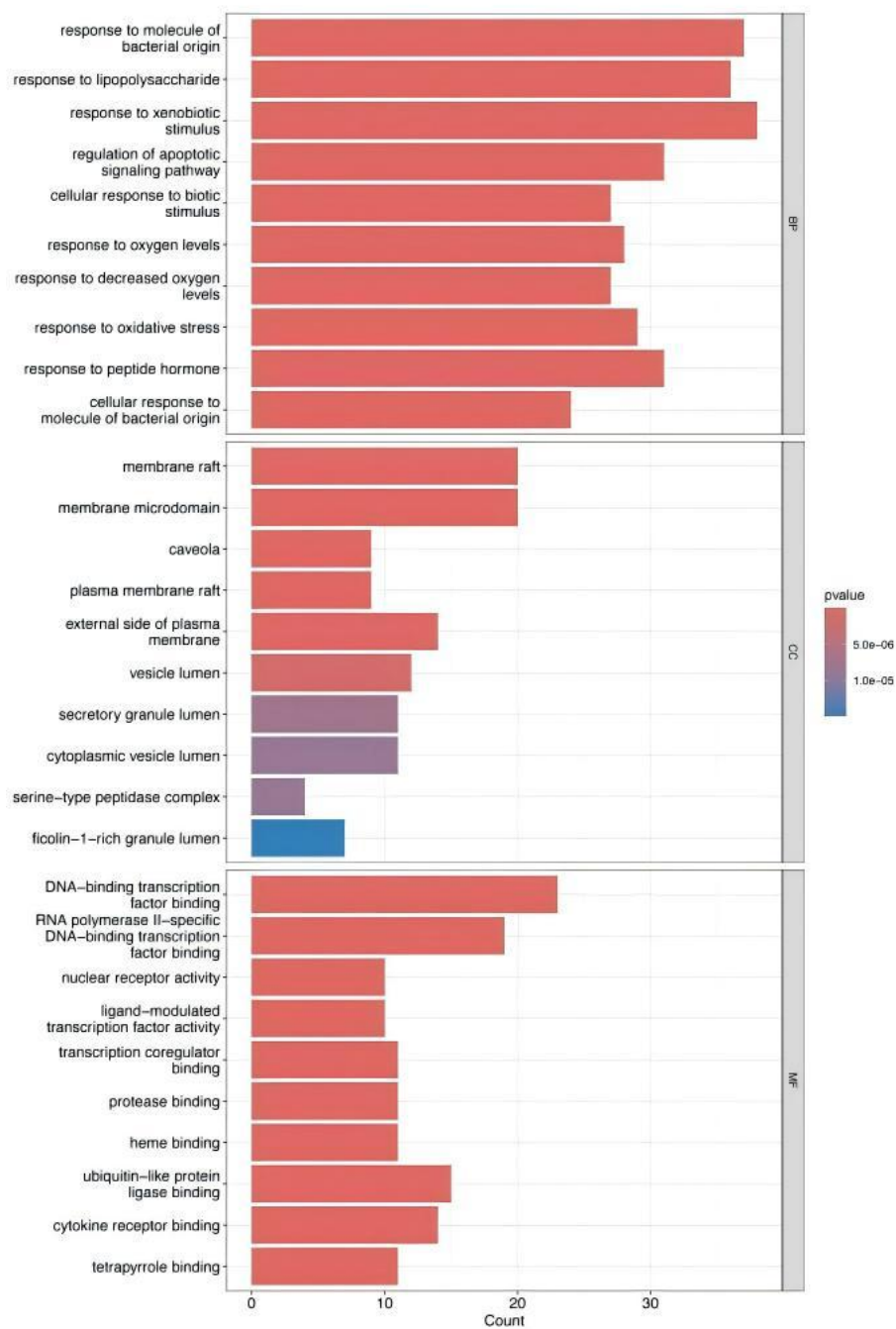


Figure 5. GO analysis of potential therapeutic targets of *Achyranthes bidentata*.

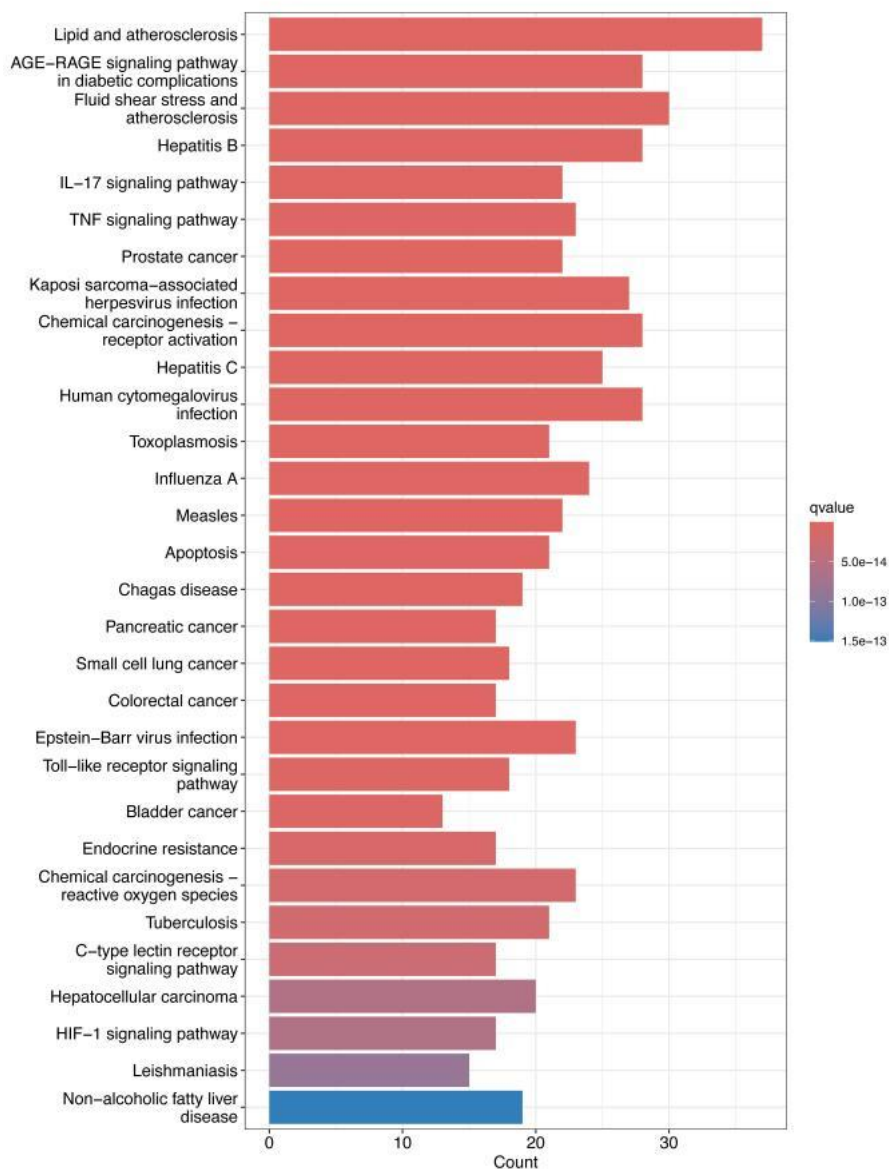


Figure 6. KEGG pathway enrichment analysis of potential therapeutic targets of *Achyranthes bidentata*.

4. Discussion

As a high-incidence cerebrovascular disease, stroke has clinical characteristics of sudden onset and severe condition. When stroke occurs, the structural integrity of the blood-brain barrier is damaged, leading to the leakage of blood components and causing neuronal dysfunction, accompanied by morphological changes in astrocytes. Existing studies have shown that intervention with natural drugs can effectively protect astrocytes from injury, inhibit their pathological activation, and reduce the levels of inflammatory mediators, providing a potential therapeutic strategy for neurofunctional repair after ischemic stroke. Other studies have indicated that active components of traditional Chinese medicine can influence the pathological progression of ischemic stroke by regulating the autophagy process, thereby improving blood perfusion in ischemic areas and promoting neurofunctional recovery.

Traditional medicine theory indicates that internal damp turbidity condensation can form phlegm-fluid retention, and poor blood circulation produces stasis; the two can transform into each other and may combine. If they block the brain meridians, stroke is easily induced.

The findings of this study indicate that the primary active compounds in *Achyranthes bidentata* responsible for treating stroke are likely quercetin, kaempferol, and wogonin. Some studies have shown that quercetin can improve the learning and memory ability of ischemic stroke mice by upregulating the phosphatidylinositol 3-kinase/protein kinase B/mammalian target of rapamycin pathway, enhance brain function, alleviate brain inflammatory response and oxidative stress response, thereby promoting the recovery of ischemic stroke mice [6]. Kaempferol and quercetin exhibit similar pharmacological effects, which can alleviate brain nerve damage in rats subjected to ischemia-reperfusion by suppressing oxidative stress and cell apoptosis, as well as protecting the blood-brain barrier [7]. Wogonin is a natural flavonoid compound. Pharmacological experiments show that it has antioxidant, vascular protective, anti-inflammatory, and neuroprotective effects. It can also inhibit the production of inflammatory mediators and neuronal apoptosis [8,9].

According to the core target analysis of the PPI network in this study, *Achyranthes bidentata* may treat stroke by acting on TP53, MAPK1, and AKT1. TP53 is involved in multiple metabolic pathways, including lipid metabolism. Additionally, some researchers have identified TP53 as a genetic marker that can help predict stroke prognosis [10]. MAPK1 is a member of the MAPK signaling pathway and has a close association with cell apoptosis and inflammatory responses in ischemic stroke. Inhibition of MAPK1 has been shown to effectively alleviate the inflammatory response induced by ischemia-reperfusion injury [11]. AKT belongs to protein kinase B and plays a key role in cell survival and apoptosis. Studies have shown that early activation of AKT can reduce neural injury in the rat brain after ischemia-reperfusion, improve hypoxia, and also has neuroprotective effects. Deficiency of the AKT1 protein can lead to abnormalities in blood lipid metabolism, among other effects [12].

5. Conclusions

In summary, this study reveals that *Achyranthes bidentata* treats stroke through multiple components, multiple targets, and multiple pathways. It predicts that its potential mechanism may involve active ingredients such as quercetin, kaempferol, and wogonin regulating key genes including TP53, MAPK3, and AKT1F, achieving treatment via pathways like Lipid and atherosclerosis, AGE-RAGE signaling pathway in diabetic complications, and Fluid shear stress and atherosclerosis. This provides a theoretical basis for further exploring the mechanism and clinical application of *Achyranthes bidentata* in treating stroke.

Conflicts of Interest

The authors declare that there is no conflict of interest regarding the publication of this article.

Data Availability

Data underlying the study's conclusions are available from the corresponding author upon request.

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References

- [1] Barthels, D.; H. Das, Current advances in ischemic stroke research and therapies. *Biochim Biophys Acta Mol Basis Dis*, 2020, 1866(4), 165260.
- [2] Diener, H.C. and G.J. Hankey, Primary and Secondary Prevention of Ischemic Stroke and Cerebral Hemorrhage: JACC Focus Seminar. *J Am Coll Cardiol*, 2020, 75(15), 1804-1818.
- [3] Tu, W.J.; L.D. Wang, China stroke surveillance report 2021. *Mil Med Res*, 2023, 10(1), 33.
- [4] Zhai, Z., et al. Progress on traditional Chinese medicine in treatment of ischemic stroke via the gut-brain axis. *Biomed Pharmacother*, 2023, 157, 114056.
- [5] Shen, H., et al. Intravenous administration of achyranthes bidentata polypeptides supports recovery from experimental ischemic stroke in vivo. *PLoS One*, 2013, 8(2), e57055.
- [6] Li, L. et al. Quercetin improves cerebral ischemia/reperfusion injury by promoting microglia/macrophages M2 polarization via regulating PI3K/Akt/NF- κ B signaling pathway. *Biomed Pharmacother*, 2023, 168, 115653.
- [7] Wang, J. et al., Kaempferol Protects Against Cerebral Ischemia Reperfusion Injury Through Intervening Oxidative and Inflammatory Stress Induced Apoptosis. *Front Pharmacol*, 2020, 11, 424.
- [8] Banik, K., et al. Wogonin and its analogs for the prevention and treatment of cancer: A systematic review. *Phytother Res*, 2022, 36(5), 1854-1883.
- [9] Feng, Y., et al. Protective role of wogonin following traumatic brain injury by reducing oxidative stress and apoptosis via the PI3K/Nrf2/HO-1 pathway. *Int J Mol Med*, 2022. 49(4).
- [10] Xie, W. et al. HMGB1-triggered inflammation inhibition of notoginseng leaf triterpenes against cerebral ischemia and reperfusion injury via MAPK and NF- κ B signaling pathways. *Biomolecules*, 2019, 9(10).
- [11] Vikman, P., et al. Cerebral ischemia induces transcription of inflammatory and extracellular-matrix-related genes in rat cerebral arteries. *Exp Brain Res*, 2007, 183(4), 499-510.
- [12] Weiss, H.R. et al. Akt activation improves microregional oxygen supply/consumption balance after cerebral ischemia-reperfusion. *Brain Res*, 2018, 1683, 48-54.



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