Rhubarb Attenuates Cerebral Edema via Inhibition of the Extracellular Signal-regulated Kinase Pathway Following Traumatic Brain Injury in Rats

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Submitted: 22-06-2017 Revised: 05-07-2017 Published: 20-02-2018

ABSTRACT

Background: Rhubarb is a traditional Chinese medicine for treating traumatic brain injury (TBI). Purpose: The purpose of this study is to elucidate the potential mechanism of rhubarb by suppressing extracellular signal-regulated kinase (ERK) to ameliorate brain edema. Materials and Methods: Sprague-Dawley rats were separated into four groups at random. One group received 3 g/kg rhubarb, and another group received 12 g/kg rhubarb, and the vehicle group and sham group were administered the same dose of saline solution. The blood-brain barrier disruption and edema were detected by Evans blue extravasation and water content, respectively. ERK, Matrix metalloproteinase 9 (MMP-9), and zona occluden-1 (ZO-1) in the damaged tissue were measured by western blot analysis and quantitative real-time polymerase chain reaction. Results: Rhubarb attenuated the brain edema after TBI, especially at the dose of 12 g/kg. Rhubarb significantly suppressed ERK, down-regulated MMP-9, and up-regulated ZO-1. Rhubarb might be a prospective therapeutic regimen to decrease edema in TBI. Conclusions: Rhubarb alleviates the edema by restraining the ERK signaling pathway. Our results contribute to the validation of the traditional use of rhubarb in the treatment of TBI and its mechanism.

Key words: Brain edema, extracellular signal-regulated kinase, rhubarb, traditional Chinese medicine, traumatic brain injury

SUMMARY

• The aim of this study was to explore the potential mechanism of rhubarb by suppressing extracellular signal-regulated kinase to ameliorate brain edema. Results: Rhubarb ameliorates edema caused by traumatic brain injury by inhibiting the ERK/Matrix metalloproteinase 9/zonula occluden-1 signaling pathway.

INTRODUCTION

Traumatic brain injury (TBI) is a disastrous condition related to substantial mortality and lifelong disability.[1] TBI has tremendous effects on both public health and the economic burden, and it carries high rates of mortality and disability in the United States,[2] with emergency hospitalization.[3] The economic burden is approximately up to more than 235,000 require hospitalization.[3] The economic burden is approximately up to more than $37.8 billion.[4] Thus, the discovery of preventive and effective drugs remains to be drastically explored.

Brain edema is a crucial factor of brain injuries to the central nervous system.[5] Following TBI, the initial mechanical insult of TBI involves breaking down the blood-brain barrier (BBB). The breakdown of the BBB leads to leakage of plasma substances and proteins out of the bloodstream and their deposition in the subendothelial matrix and interstitium. The above alterations cause the accumulation of plasma proteins in the subendothelial matrix, further resulting in brain edema. Cerebral edema may be responsible for as many as half of the mortalities in TBI.[6] Hence, alleviating cerebral edema is a contributing factor to neurologic recovery.[7]

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Cite this article as: Yang Z, Fan R, Sun P, Cui H, Peng W, Luo J, et al. Rhubarb attenuates cerebral edema via inhibition of the extracellular signal-regulated kinase pathway following traumatic brain injury in rats. Phcog Mag 2018;14:134-9.
Several medicines, including ethanol, carbamylated erythropoietin and astaxanthin, are used to attenuate cerebral edema after TBI. Current medical approaches to effectively alleviate cerebral edema after TBI is not available. The reason is that due to the complex pathogenesis of TBI, “one-compound, one-disease” based drugs fail to alleviate brain edema after TBI. Scientists and doctors tend to focus on traditional Chinese medicine (TCM) for the treatment of TBI because of its multiple-targets. The TCM herb Rheum officinale Baill (Rhubarb, a member of the Polygonaceae family), the dried roots and rhizomes, has been used for thousands of years of ancient medicine. It is recorded in Shennong’s Classic of Material Medical (Shen Nong Ben Cao Jing). Rhubarb has several pharmacological effects, including antiinflammatory, antibacterial, purgative, and anticancer properties. Furthermore, a previous study demonstrates that rhubarb is highly efficient in treating patients with TBI by its multitargets effects. However, the molecular mechanism of rhubarb in the alleviation of brain edema is still unknown. Extracellular signal-regulated kinase (ERK) is a member of mitogen-activated protein kinases (MAPKs), which is activated after TBI. Matrix metalloproteinase 9 (MMP-9) is downstream of ERK and functions to degrade the major components of the basal lamina, such as the tight junctions, which cause BBB disruption after TBI, leading to edema. Tight junction proteins, such as zonula occluden-1 (ZO-1), are a symbol of the integrity of the BBB that essentially contribute to its structural inviolacy. A change in tight junction protein assembly may contribute to the loss of the BBB integrity and BBB breakdown. A study showed that the prohibition of the ERK pathway ameliorates cortical damage after cerebral trauma. Therefore, inhibiting the ERK signaling pathway is a key to alleviating cerebral edema. Whether rhubarb suppresses the ERK signaling pathways to ameliorate the cerebral edema remains unclear.

The present study aimed to elucidate whether rhubarb relieved brain edema through the downregulation of MMP-9 and the upregulation of ZO-1 by inhibiting the ERK signaling pathway in a rat model of TBI. This research helps to provide a promising herbal drug to treat TBI.

**MATERIALS AND METHODS**

**Experimental animals**

Adult male Sprague-Dawley rats, weighing 220-300 g, were used in this study and were purchased from the Laboratory Animal Center of Central South University. The animals were housed under controlled conditions (temperature at 25℃, 65% ±5% relatively humidity, and 12 h: 12 h light/dark cycle). All the animal experimental protocols conducted in this study were approved by the Xiangya Hospital, Central South University (Changsha, China) and were put into effect according to Institutional Guidelines of the Animal Care and Use Committee.

**Controlled cortical impact model**

The rats were anesthetized with 3% pentobarbital sodium (50 mg/kg), and the rats were mounted in a stereotaxic frame. Following a midline incision, the skin and temporal muscles were reflected to expose the skull. A craniotomy, with the diameter of 4.0 mm, was performed using dental bur at the left. Briefly, a controlled cortical impact (CCI) device (TBI0310, precision systems and instrumentation, Fairfax Station, VA) was used to impact the brain (tip diameter, 4.0 mm; cortical contusion depth, 5.0 mm; impact velocity, 6.0 m/s; dwell time, 50.0 ms). After injury, the skin incision was closed with nylon sutures. The sham group was only subject to craniotomy. The rats were placed on a heating pad during surgery. At the end of the procedure, the animals were closely monitored until the recovery from anesthesia was complete.

**Experimental groups and the administration of drugs**

Eighty rats were randomly divided into four groups in a blinded manner, including the sham (rats underwent the CCI procedure without impacting the cortex and were gavaged with 0.9% saline, n = 20), vehicle (rats underwent the CCI procedure and received the same dose of saline, n = 20), rhubarb 3 g/kg in distilled water (CCI rats and received rhubarb 3 g/kg, n = 20), and rhubarb 12 g/kg in distilled water (CCI rats and received rhubarb 12 g/kg, n = 20). The drugs were administered by gavage after post-TBI recovery. The investigators assessed all of the study outcomes, performed the calculations, and conducted the analyses. All the rats were calculated in the final blind data analysis expect for those that died before the end of the study.

**Preparation of rhubarb**

The Rheum officinale Baill (dried roots and rhizomes, voucher specimen No. 20140624, Gansu, China) was purchased from the pharmacy of the Xiangya Hospital, Central South University. An herbal medicine botanist, Professor Suiyu Hu (Department of Herbal Medicine of Central South University in China), authenticated the plant. The rhubarb was boiled twice in distilled water (1:12, v/w) for 30 min.

**Brain water content**

The brain water content was used on behalf of edema, which forms as a consequence of the BBB breakdown. After decapitating the rats (n = 5 in each group), the brains were removed. The injured hemisphere was weighed (wet weight), dried at 100°C for 24 h, and reweighed (dry weight). The water content was determined by the equation [{wet weight – dry weight} / wet weight] ×100%. 

**Evaluation of the blood–brain barrier permeability**

Evans blue (EB) extravasation was used to investigate the BBB disruption. In short, the EB dye (2 ml/kg in 20% saline) was administered through the tail vein and was allowed to circulate for 2 h. Two hours later, the animals were anesthetized and perfused with saline to remove intravascular EB dye. The rats (n = 5 in each group) were then decapitated, and the injured cortical tissues were harvested. Each tissue sample was weighed, homogenized in a 50% trichloroacetic acid solution and centrifuged at 10,000 rpm for 10 min. The supernatant was diluted with ethanol (1:3). The absorbance of each supernatant for the EB dye was measured at 620 nm using a spectrophotometer.

**Immunohistochemistry**

The rats (n = 5 in each group) were anesthetized with 3% pentobarbital sodium (50 mg/kg), and then, the rats were decapitated, and the brain was removed and stored in 4% paraformaldehyde until processing. Frozen areas (10 μm) of the injured cortex were brought to room temperature and were then incubated in 3% H₂O₂ for 10 min. After washing three times in phosphate buffer saline (PBS) for 5 min each at room temperature, 5% normal donkey serum was used to block the nonspecific binding. Immunostaining was performed using primary antibodies (1:400, Abcam ab59720, USA) specific for ZO-1 and antibodies for MMP-9 (1:400, Epitomics 2551-1, USA) at 4°C overnight. Next, staining with biotin-labeled secondary antibodies for 120 min was performed. Then, the avidin-biotin-peroxidase complex (1:100, Sigma, USA) was used at 37°C for 1 h. Diaminobenzidine (Boster Biotech Co. Wuhan, China) was applied to visualize the immunoreactivity. Under the light microscope, positive staining (brown yellow) was located for ZO-1 and MMP-9. The images were under a magnification of 400 × by randomly choosing 10 microscopic fields from each group, and the...
digital software Image-ProPlus 5.0 (Media Cybernetics, USA) was used to automatically detect the integral optical density of each group.

**Quantitative real-time polymerase chain reaction analysis**

The rats ($n = 5$ in each group) were anesthetized with $3\%$ pentobarbital sodium ($50$ mg/kg), underwent cardiac perfusion with ice-cold saline. Cortex was dissected from the brain tissue using a fine-straight and a fine-angled dissecting forceps and immediately transferred to liquid nitrogen. For total RNA isolation from the ipsilateral cerebral cortices that had been treated with PBS using the Trizol reagent (Invitrogen, USA), the cortices were homogenized in $1$ ml of Trizol (Invitrogen, USA), and the total RNA was isolated using the manufacturer's instructions. Spectrophotometry was used to confirm the purity. The green polymerase chain reaction (PCR) kit (Fermentas, USA) and gene-specific primers were to quantify the mRNA on a Bio-Rad C × 96 Detection System (Bio-Rad, USA). cDNA served as a template for quantitative real-time RT-PCR. $\beta$-actin was used as an internal control to normalize the of gene expression levels. ZO-1 and MMP-9 were amplified with the specific primers presented in Table 1.

**Western blot analysis**

The rats ($n = 5$ in each group) were anesthetized with $3\%$ pentobarbital sodium ($50$ mg/kg), and perfused transcardially with ice-cold saline. The rats ($n = 5$ in each group) were anesthetized with $3\%$ pentobarbital sodium ($50$ mg/kg), underwent cardiac perfusion with ice-cold saline. Cortex was dissected from the brain tissue using a fine-straight and a fine-angled dissecting forceps and immediately transferred to liquid nitrogen. For total RNA isolation from the ipsilateral cerebral cortices that had been treated with PBS using the Trizol reagent (Invitrogen, USA), the cortices were homogenized in $1$ ml of Trizol (Invitrogen, USA), and the total RNA was isolated using the manufacturer's instructions. Spectrophotometry was used to confirm the purity. The green polymerase chain reaction (PCR) kit (Fermentas, USA) and gene-specific primers were to quantify the mRNA on a Bio-Rad C × 96 Detection System (Bio-Rad, USA). cDNA served as a template for quantitative real-time RT-PCR. $\beta$-actin was used as an internal control to normalize the of gene expression levels. ZO-1 and MMP-9 were amplified with the specific primers presented in Table 1. The thermal cycling began with a $2$ min incubation at $50^\circ$C followed by a $10$ min denaturation step at $95^\circ$C and $40$ cycles at $95^\circ$C for $10$ sand $59^\circ$C for $50$ s. The comparative threshold cycle (Ct) method was used to determine the relative quantities of the candidate genes and $\beta$-actin mRNA.

**Table 1: Reverse transcription polymerase chain reaction primer sequences**

| Gene   | Primers       | Sequences                              | Length |
|--------|---------------|----------------------------------------|--------|
| MMP-9  | Forward       | 5'-GCAAACCTGCGTATTTCCAT-3'             | 76 bp  |
|        | Reverse       | 5'-CCATCCGGAGCGACCTTGTAG-3'            |        |
| ZO-1   | Forward       | 5'-CATCGCTCGTTGCGACCGATTG-3'           | 201 bp |
|        | Reverse       | 5'-CATCGCTCGTTGCGACCGATTG-3'           |        |
| Actin  | Forward       | 5'-TAATGTCACGCACGATTGCC-3'             | 107 bp |
|        | Reverse       | 5'-TAATGTCACGCACGATTGCC-3'             |        |

**Rhubarb alleviates trauma-induced phospho-extracellular signal-regulated kinase and extracellular signal-regulated kinase levels**

The expression of p-ERK protein was dramatically increased in the CCI model, EB extravasation was used to investigate the BBB disruption. Similarly, rats treated with rhubarb showed a significantly decreased BBB disruption in a dose-dependent manner. We observed a significant difference in the $3$ g/kg and $12$ g/kg rhubarb-treated groups compared with the vehicle-treated group [Figure 1b].

**Figure 1:** (a) Compared the brain water content (%) after traumatic brain injury in rats. The water content increased after traumatic brain injury and treated with rhubarb alleviated it. (b) Evans Blue is used to detect the blood–brain barrier permeability. Posttraumatic brain injury leads to blood–brain barrier disruption, the uptake of rhubarb remits the blood–brain barrier permeability. The data are represented as the mean ± standard deviation. *$P < 0.05$, $n = 5$ in each group.
treatment (3 g/kg and 12 g/kg) inhibited the TBI-induced activation of the p-ERK and ERK [Figure 2a]. We observed a significant difference in the 3 g/kg and 12 g/kg rhubarb-treated groups compared with the vehicle-treated group (P < 0.05), [Figure 2a].

**Figure 2:** Expression of the protein in the brain tissue ipsilateral to the injury after treatment (n = 5). (a) p-extracellular signal-regulated kinase and extracellular signal-regulated kinase. (b) Matrix metalloproteinase 9. (c) zonula occluden-1. The data are represented as the mean ± standard deviation. *: P < 0.05, **: P < 0.01, n = 5 in each group.

**Figure 3:** Comparison of (a) matrix metalloproteinase 9 and (b) zonula occluden-1 expressions after traumatic brain injury. (a) Separate treatments with 3 g/kg rhubarb and 12 g/kg rhubarb significantly reduce the matrix metalloproteinase 9. (b) Compared with the vehicle, treating with 12 g/kg rhubarb significantly increased the zonula occluden-1 expression. The data are represented as the mean ± standard deviation. **: P < 0.01, *: P < 0.05, n = 5 in each group.
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Rhubarb decreases the upregulation of matrix metalloproteinase 9 and reduces the degradation of the tight junction protein ZO-1

The results of the immunohistochemical examination, real-time PCR, and western blot are shown in Figure 3a and b, Figure 4a and b, and Figure 2b and c. In the vehicle group compared to the sham group, we observed the activation of MMP-9 and the reduction of ZO-1. The rhubarb treatment decreased MMP-9 expression and was accompanied by increased ZO-1 expression. Furthermore, the Western blot and real-time PCR revealed that rhubarb downregulated the expression levels of the MMP-9 mRNA [Figure 4a] and protein [Figure 2b] and was accompanied by elevated the expression levels of the ZO-1 mRNA [Figure 4b] and protein [Figure 2c].

DISCUSSION

This study revealed that rhubarb significantly decreased the extent of cerebral edema and the permeability of the BBB in the CCI model. Moreover, a suppression of ERK and MMP-9, to activate ZO-1, was observed. The results showed that rhubarb provided a significant neuroprotection to ameliorate brain edema formation through inhibiting the ERK signaling pathway. This research provided a potential molecular mechanism of rhubarb in the treatment of TBI.

The ERK pathway, which regulates the activity of many transcriptional factors associated with the proliferation of various cells, is activated in lesions in regions of selective vulnerability after TBI.[22] Raf/mitogen extracellular kinase (MEK)/ERK is the canonical pathway to activate ERK, in which the β-Raf activation of MAPK (MEK) that then phosphorylates and activates ERK.[16] MMP-9 is one of the inducible enzymes[23] of MMPs, which degrade tight junctions in the brain.[25] MMP-9 activity is significantly elevated in TBI.[24] Several studies demonstrate that MMP-9 proteolytic activity is associated with brain edema.[25‑27] ZO-1 is present in brain endothelial tight junctions, which are the main structural barrier proteins.[23] Previously, studies showed that inhibit ERK reduces MMP-9 levels and increases ZO-1 levels, which then attenuates traumatic brain edema.[15] Until now, it was not reported that the TCM rhubarb, by restrained ERK, decreasing MMP-9 and increasing ZO-1, alleviates edema.

As presented in Figure 5, the brain is sensitive to brain edema following TBI, which may be successfully treated on the condition of a timely beginning. Our previous study indicated rhubarb protects the BBB following TBI through the NADPH oxidase/ROS/ERK/MMP-9/ZO-1 signaling pathway.[28] The figure showed that rhubarb lowered the MMP-9 level and raised ZO-1 expression. Moreover, it is necessary to analyze the mechanism of how the brain edema is alleviated after TBI through MMP-9 inhibition and ZO-1 elevation, especially for the ERK signaling pathway. Therefore, we assessed the influence of rhubarb on the ERK/MMP-9/ZO-1 signaling pathway. The data suggested that rhubarb stopped the effects of ERK, which subsequently prohibited MMP-9 and boosted ZO-1. The results presented that rhubarb might be used as a potential therapeutic agent to attenuate brain edema in the treatment of TBI.

This study has several limitations. Although rhubarb alleviate edemases, it is unclear that the absorbed bioactive compositions derived from rhubarb exert are protective functions in the brain. Furthermore, it is unknown the manner by which the activated ERK/MMP-9/ZO-1 signaling pathway was suppressed after TBI is unknown. More efforts are needed to elucidate the mechanisms underlying these symptoms.

CONCLUSIONS

Rhubarb exerts neuroprotective effects by ameliorating edema caused by TBI through inhibiting the ERK/MMP-9/ZO-1 signaling pathway.[15] Therefore, rhubarb has potential as a promising neuroprotective candidate for TBI.
Acknowledgements

The project was supported by the Natural Science Foundation of the Hunan Province, China (Grant No. 2017jj2376). The author thanks Rong Fan, Peng Sun, Hanjin Cui, and Weijun Peng for helping with experiments, Jiekun Luo, Chunhu Zhang, Xingui Xiong, and Wei Huang for technical guidance, and Wei Liu, for the experimental design, article writing, and statistical analysis, Wei Liu also support the funding.

Financial support and sponsorship

The project was supported by the Natural Science Foundation of the Hunan Province, China (Grant No. 2017jj2376).

Conflicts of interest

There are no conflicts of interest.

REFERENCES

1. Nichol A, French C, Little L, Haddad D, Presneill J, Arabi Y, et al. Erythropoietin in traumatic brain injury (EPO-TBI): A double-blind randomised controlled trial. Lancet 2015;386:2499-506.
2. Wang W, Li H, Yu J, Hong M, Zhou J, Zhu L, et al. Protective effects of chinese herbal medicine rhizoma drynariae in rats after traumatic brain injury and identification of active compound. Mol Neurobiol 2016;53:4809-20.
3. Lozano D, Gonzales-Porillo GS, Acosta S, de la Pena I, Tajni N, Kaneko Y, et al. Neuroinflammatory responses to traumatic brain injury: Etiology, clinical consequences, and therapeutic opportunities. Neuropsychiatr Dis Treat 2015;11:97-108.
4. Luo CL, Li QQ, Chen XP, Zhang XM, Li LL, Li BX, et al. Lipoxin A4 attenuates brain damage and downregulates the production of pro-inflammatory cytokines and phosphorylated mitogen-activated protein kinases in a mouse model of traumatic brain injury. Brain Res 2013;1502:1-10.
5. Donkin JJ, Vink R. Mechanisms of cerebral edema in traumatic brain injury: Therapeutic developments. Curr Opin Neurol 2010;23:293-9.
6. Rossi JL, Todd T, Daniels Z, Bazan NG, Belaya L. Interferon-stimulated gene 15 upregulation precedes the development of blood-brain barrier disruption and cerebral edema after traumatic brain injury in young mice. J Neurotrauma 2015;32:1101-8.
7. Wang T, Chou DY, Ding JY, Fredrickson V, Peng C, Schaffer S, et al. Reduction of brain edema and expression of aquaporins with acute ethanol treatment after traumatic brain injury. J Neurosurg 2013;118:390-6.
8. Bouza R, Francony G, Thomas S, Valable S, Mauconduit F, Favre MC, et al. Reduced brain edema and functional deficits after treatment of diffuse traumatic brain injury by carbamylated erythropoietin derivative. Crit Care Med 2011;39:2099-105.
9. Zhang M, Cui Z, Cui H, Cao Y, Zhong C, Wang Y, et al. Astaxanthin alleviates cerebral edema by modulating NKCC1 and AQP4 expression after traumatic brain injury in mice. BMC Neurosci 2016;17:60.
10. Hakon J, Ruscher K, Rommer B, Tomasevic G. Preservation of the blood brain barrier and cortical neuronal tissue by liraglutide, a long acting glucagon-like-1 analogue, after experimental traumatic brain injury. PLoS One 2015;10:e0120074.
11. Lu K, Zhang C, Wu W, Zhou M, Tang Y, Peng Y, et al. Rhubarb extract has a protective role against radiation-induced brain injury and neuronal cell apoptosis. Mol Med Rep 2015;12:2689-94.
12. Wang Y, Huang X, Liang QH, Fan R, Qin F, Guo Y, et al. A strategy for detecting absorbed bioactive compounds for quality control in the water extract of rhubarb by ultra performance liquid chromatography with photodiode array detector. Chin J Integr Med 2012;18:690-8.
13. Wang Y, Fan R, Luo J, Tang T, Xing Z, Xia Z, et al. An ultra high performance liquid chromatography with tandem mass spectrometry method for plasma and cerebrospinal fluid pharmacokinetics of rhein in patients with traumatic brain injury after administration of rhubarb decoction. J Sep Sci 2015;38:1100-8.
14. Gu J, Zhang X, Fei Z, Wen A, Qin S, Yi S, et al. Rhubarb extracts in treating complications of severe cerebral injury. Chin Med J (Engl) 2000;113:529-31.
15. Mori T, Wang X, Aoki T, Lo EH. Downregulation of matrix metalloproteinase-9 and attenuation of edema via inhibition of ERK mitogen activated protein kinase in traumatic brain injury. J Neurotrauma 2002;19:1411-9.
16. Atkins CM, Falo MC, Alonso OE, Bramlett HM, Dietrich WD. Deficits in ERK and CREB activation in the hippocampus after traumatic brain injury. Neurosci Lett 2009;459:52-6.
17. Raghupathi R, Mud JR, Fulp CT, Pittman RN, McIntosh TK. Acute activation of mitogen-activated protein kinases following traumatic brain injury in the rat: Implications for posttraumatic cell death. Exp Neurol 2003;183:438-48.
18. Shao AW, Wu HJ, Chen S, Ammar AB, Zhang JM, Hong Y. Resveratrol attenuates early brain injury after subarachnoid hemorrhage through inhibition of NF-kappaB-dependent inflammatory/MMP-9 pathway. CNS Neurosci Ther 2014;20:182-6.
19. Chen CC, Hung TH, Lee CY, Wang LF, Wu CH, Ke CH, et al. Berberine protects against neuronal damage via suppression of glia-mediated inflammation in traumatic brain injury. PLoS One 2014;9:e115694.
20. Luh C, Kuhlmann CR, Ackermann B, Timaru-Kast R, Luhmann HJ, Behl C, et al. Inhibition of myosin light chain kinase reduces brain edema formation after traumatic brain injury. J Neurochem 2010;112:1025-25.
21. Mori T, Wang X, Jung JC, Sumii T, Singhal AB, Fini ME, et al. Mitogen-activated protein kinase inhibition in traumatic brain injury: in vitro and in vivo effects. J Cereb Blood Flow Metab 2002;22:444-52.
22. Otani N, Navashin H, Fukushima S, Osogawa H, Ohsumi A, Toyocka T, et al. Role of the activated extracellular signal-regulated kinase pathway on histological and behavioral outcome after traumatic brain injury in rats. J Clin Neurosci 2007;14:42-8.
23. Yang Y, Rosenberg GA. Matrix metalloproteinases as therapeutic targets for stroke. Brain Res 2015;1623:30-8.
24. Abdul-Muneer PM, Pfister BJ, Haasch J, Chandra N. Role of matrix metalloproteinases in the pathogenesis of traumatic brain injury. Mol Neurobiol 2016;53:6106-23.
25. Cui J, Chen S, Zhang C, Meng F, Wu W, Hu R, et al. Inhibition of MMP-9 by a selective gelatinase inhibitor protects neurovascular from embolic focal cerebral ischemia. Mol Neurodegener 2012;7:21.
26. Gasche Y, Fujimura M, Morita-Fujimura Y, Copin JC, Kawase M, Massengale J, et al. Early appearance of activated matrix metalloproteinase-9 after focal cerebral ischemia in mice: A possible role in blood-brain barrier dysfunction. J Cereb Blood Flow Metab 1999;19:1020-8.
27. Wang J, Torika SE. Neuroprotection by inhibition of matrix metalloproteinases in a mouse model of intracerebral haemorrhage. Brain 2005;128:1623-32.
28. Wang Y, Fan X, Tang T, Fan R, Zhang C, Huang Z, et al. Rhein and rhubarb similarly protect the blood brain barrier after experimental traumatic brain injury via gp91phox subunit of NADPH oxidase/ROS/ERK/MMP-9 signaling pathway. Sci Rep 2016;6:37098.