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ORIGINAL ARTICLE

The influence of stachydrine hydrochloride on the reperfusion model of mice with repetitive cerebral ischemia

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KEYWORDS

Stachydrine hydrochloride;
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Abstract To study the influence of stachydrine hydrochloride on the inflammatory cytokines and tissue morphology of the re-perfusion model of mice with repetitive cerebral ischemia and probe into the protection mechanism of stachydrine hydrochloride for cerebral ischemia reperfusion impairment. Build a repetitive cerebral ischemia reperfusion model by first blocking the common carotid artery on both sides for 10 min, then resuming perfusion for 10 min and then blocking the common carotid artery on both sides again for 10 min. Before the operation, all the mice in the Nimodipine group, and the big, medium and small stachydrine hydrochloride dose groups were given corresponding gastric perfusion, the mice in the sham operation group and the modeled groups were at the same time given 0.5% sodium carboxymethyl cellulose for gastric perfusion of the same volume. The medicine was fed daily for 7 consecutive days. The model was built 1 h after the last feed and the perfusion continued for 24 h after the operation. Then the death rate of the mice was calculated. The mouse brains were taken out to test the ICAM-1 level and the TNF-α level, and the serum was taken out to test the NSE level and the MPO level. The tissue morphology changes were also observed. All the repetitive cerebral ischemia reperfusion models were successfully duplicated. The stachydrine hydrochloride in all the dose groups significantly reduced the death rates of big and small mice, reduced the level of ICAM-1 and the level of TNF-α in the brain tissues and the NSE level and the MPO level in the serum, significantly alleviating the pathological impairment in the hippocampus. Stachydrine hydrochloride can significantly reduce the

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death rate of mice, improve the pathological changes in the hippocampus, inhibit inflammatory reactions after ischemia, thus reducing the re-perfusion impairment after cerebral ischemia.

1. Introduction

Stachydrine hydrochloride is an important part of the pharmacological basis for alkaloids in the *Leonurus japonicus* Houtt - motherwort. The motherwort tastes spicy and slightly bittersweet. The property slightly, it can be used for clearing heat and detoxification. Stachydrine hydrochloride is also called Proline Betaine or N-Dimethyl proline and is the simplest pyrrole alkaloid. Its basis structure is L-stachydrine and its molecular weight M = 143.18 and the pH = 7. The Chinese Pharmacopoeia 2015 has already designated the concentration of stachydrine hydrochloride as the standard for testing dry motherwort – the concentration of stachydrine hydrochloride must not be below 0.50%. The pyrrole rings in their chemical structure have very good medical effects. Modern pharmacological studies have shown that the alkaloids in motherwort have extensive pharmacological activities in treating cardiovascular and cerebral diseases and resisting inflammations. Cerebral ischemia is a common disease among old people and the pathological mechanism of impairment caused by cerebral ischemia is very complex, which includes many factors, such as the emergence of inflammatory media, exhaustion of energy (Liu, 2016), the emergence of radicals, and the activation of apoptosis pathway. Early studies have found that motherwort, having the capacity to clear heat, detoxify, activate blood and remove blood stasis, can be used in alleviating the impairment caused by cerebral ischemia. "Activate blood and remove blood stasis" is commonly used in clinical treatment of impairment caused by cerebral ischemia and the effect is significant. "Clear heat and detoxify" is a new viewpoint in alleviating such impairment put forward by traditional Chinese medicine researchers.

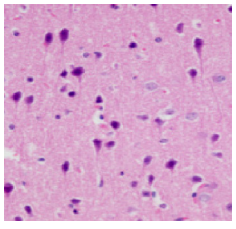
2. Materials and methods

2.1. Drugs and reagents

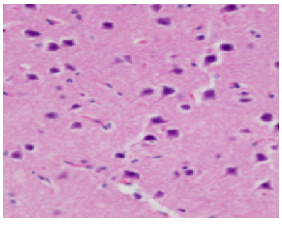
Stachydrine hydrochloride, was provided by the Chemical Lab of Henan University of Traditional Chinese Medicine, concentration > 9%, batch No: 20091212; Nimodipine pills, by Yabao Pharmaceutical Group Co., Ltd, batch No: 130150; penicillin sodium for injection use, by Huabei Pharmaceuticals Co., Ltd, batch No. C1206807; CMC, by Hengxing Chemical Reagent Production Co., Ltd of Tianjin, batch No.: 20120418; ICAM-1 ELISA Testing Reagent, R&D Company, batch No. 20130901A; TNF-α ELISA testing reagent box, R&D Company, batch No. 20130901A; NSE ELISA testing reagent box, R&D Company, batch No. 20130901A; MPO testing box, Nanjing Jiancheng Bio-engineering Institute, batch No. 20130914. Ultraviolet–visible spectrophotometer, provided by Shanghai Tianmei Scientific Instrument Co., Ltd, type: UV 1000; reader, BIO-RAD Company (US), type: BIORAD-68 (Tables 1 and 2).

2.2. Animals

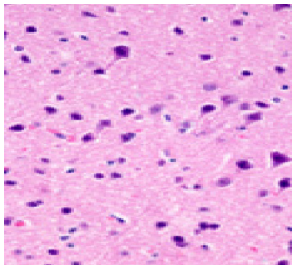
KM mice, SPF level, male, 96 in number, weighted 25–30 g, were provided by the Experiment Animal Center of Henan Province, license No: 41003100000258; lab license No.: SYXK (Henan) 2010-001 (Fig. 1).



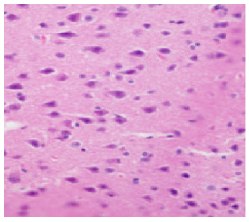
Pic.1 Sham-operation group (HE×400)



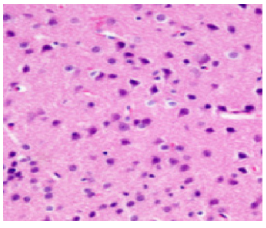
Pic.2 Model group (HE×400)



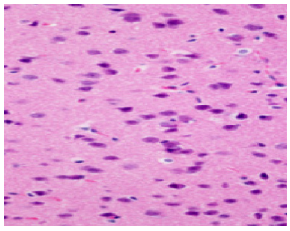
Pic.3 Nimodipine group (HE×400)



Pic.4 Big-dose group (HE×400)



Pic.5 Medium-dose group (HE×400)



Pic.6 Small-dose group (HE×400)

Figure 1 The pathological photos of the cerebral cortex of modeled mice.