

**Neuroprotective effects of ginsenoside Rb1 on primary
cultured rat cortex neural progenitor cells in
tert-butylhydroperoxide-induced oxidative injury**

By

Na Ni

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Institute of Chinese Medical Sciences

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A thesis submitted in partial fulfillment of the requirements for the degree of Master of
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Supervisor

Date

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澳門大學碩士學位論文

研究人參皂苷**Rb1**對於大鼠皮質區原代培養神經
幹細胞在叔丁基過氧化氫誘導的氧化損傷模型中
的神經保護作用

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澳門大學中華醫藥研究

University of Macau

Abstract

**Neuroprotective effects of ginsenoside Rb1 on primary cultured rat
cortex neural progenitor cells in tert-butylhydroperoxide-induced
oxidative injury**

By Na Ni

Thesis Supervisor: Dr. Huanxing Su

Chinese Medicinal Science

Ginseng, the root of *Panax ginseng* C.A. Meyer, has been used as a representative tonic to enhance bodily functions against various ailments for hundreds of years in Far Eastern countries without apparent adverse effects. Ginsenoside Rb1, one of the most active ingredients of ginseng, has been shown to possess various pharmacological activities. Here we reported that Rb1 exhibited potent neuroprotective effects against oxidative injury induced by tert-butylhydroperoxide (t-BHP). Lactate dehydrogenase (LDH) assay demonstrated that incubation with 300 μ M t-BHP for 2.5 h led to a significant cell loss of cultured rat embryonic cortex-derived neural progenitor cell (NPCs) and the cell viability was pronouncedly increased by 24 h pretreatment of 10 μ M Rb1. TUNEL staining further confirmed that pretreatment of Rb1 significantly reduced the cell apoptosis in t-BHP-induced oxidative injury. Real time PCR revealed that pretreatment with Rb1 activated Nrf2 pathway in cultured NPCs and led to an elevated expression of HO-1. The results of the present study demonstrate that Rb1 shows a potent anti-oxidative effect on

cultured NPCs by activating Nrf2 pathway.

Key words:

Ginseng, ginsenoside Rb1, neural progenitor cell, t-BHP, neuroprotective, Nrf2, HO-1



摘要

研究人參皂苷Rb1對於大鼠皮質區原代培養神經幹細胞在叔丁基過氧化氫誘導的氧化損傷模型中的神經保護作用

人參,五加科植物人參的根,在東方常被用作補藥來對抗不同疾病,增強身體機能,而且基本沒有產生副作用,已經有近百年的歷史,人參皂苷Rb1,是人參最有效的活性成分之一,具有多種藥理活性。這裏我們發現了人參皂苷Rb1對於由叔丁基過氧化氫誘導的氧化損傷具有一定的神經保護作用。LDH測試結果表明胚胎大鼠皮質區的神經幹細胞經過300 μm 的叔丁基過氧化氫處理2.5小時以後會使細胞的死亡率明顯增加,但是在經過10 μm 的Rb1處理24小時以後,細胞死亡率則明顯降低。之後TUNEL染色又對此結果進行了進一步驗證。為了探究其神經保護的作用機理,我們進行了實時熒光定量PCR的分析,結果表明經過人參皂苷Rb1預處理以後,可以激活Nrf2通路,並且會促進下遊的抗氧化基因HO-1的表達。結果表明人參皂苷Rb1可以通過激活Nrf2通路對神經幹細胞發揮抗氧化作用。

關鍵詞

人參, 人參皂苷Rb1,神經幹細胞, 叔丁基過氧化氫, 神經保護, Nrf2, HO-1

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ABBREVIATIONS

%	Percent
°C	Degree Celsius
Akt	Protein Kinase B (PKB)
ARE	Antioxidant responsive elements
BrdU	Bromodeoxyuridine
BSA	Bovine serum albumin
CAT	Catalase
CNS	Central nervous system
DAPI	4', 6-diamidino-2-phenylindole
FBS	Fetal bovine serum
HO-1	Heme oxygenase-1
LDH	Lactate dehydrogenase
MCAO	Mouse Model of Middle Cerebral Artery Occlusion
mg	Microgram
ml	Milliliter
mM	Micromole
Nrf-2	Nuclear factor (erythroid-derived 2)-like 2
NPCs	Neural progenitor cell
NQO1	NAD(P)H dehydrogenase (quinone 1)
PBS	Phosphate-buffered saline

PFA	Paraformaldehyde
PLL	Poly-l-lysine
RNS	Reactive nitrogen species
ROS	Reactive oxygen species
RT-PCR	Real time polymerase chain reaction
SOD2	Superoxide dismutase2
SVZ	Subventricular zone
TUNEL	Terminal deoxynucleotidyl transferase dUTP nick end labeling
t-BHP	Tert-Butylhydroperoxide
μm	Micromole

