

Identification of cholest-4-ene-3,6-dione as a Novel Neuroprotectant in Ischemic Stroke and Its Lipidomics

Xiaoshuai Ren^{1,*}, Feng Lin^{2,*}, Chaogang Tang^{2,*}, Yao Liu³, Guolei Liao², Jiabi Liang³, Wenji Luo³, Lei Zhang², Wenli Chen³

¹Department of Clinical Laboratory, The Fifth Affiliated Hospital of Sun Yat-Sen University, Zhuhai, Guangdong, 519000, People's Republic of China;

²Department of Cerebrovascular Disease, The Fifth Affiliated Hospital of Sun Yat-Sen University, Zhuhai, Guangdong, 519000, People's Republic of China; ³Department of Pharmacy, The Fifth Affiliated Hospital of Sun Yat-Sen University, Zhuhai, Guangdong, 519000, People's Republic of China.

*These authors contributed equally to this work.

Correspondence: Wenli Chen; Lei Zhang, Tel +15626471884; +13736209240, Email: chenwenli@mail3.sysu.edu.cn; zhang92@mail3.sysu.edu.cn

Purpose: Stroke is a leading cause of disability and death globally. However, there are few clinical drugs for stroke therapy. Novel and effective neuroprotectants are called on the way.

Methods: In this study, 93 steroids from a constructed steroidal library were randomly numbered and blindly evaluated in an L-glutamate-induced HT-22 oxidative stress model. The neuroprotective effects of 5 candidates were further investigated in potassium deprivation-induced apoptosis of cerebellar granule neurons (CGNs), D-glutamate-induced excitotoxicity of CGNs, and cortical neuron (CN) models.

Results: Interestingly, unblinding revealed that cholest-4-ene-3,6-dione (78), a cholesterol derivative, was first found to have comprehensive neuroprotective effects in all cell models. 78 administration also decreased the infarction volume and improved motor function in middle cerebral artery occlusion (MCAO) model rats. Additionally, 78 treatment decreased intercellular reactive oxygen species (ROS) and NO production in the HT-22 cell model. Finally, lipidomics and molecular docking results showed that 78 may exert its neuroprotective effects by increasing platelet-activating factor (PAF) analog 1-(9Z-pentadecenoyl)-glycero-3-phospho-choline production.

Conclusions: This study indicates that 78, a novel neuroprotectant, is a promising therapeutic candidate with comprehensive neuroprotective effects for the treatment of ischemic stroke by decreasing ROS/reactive nitrogen species (RNS) levels and increasing 1-(9Z-pentadecenoyl)-glycero-3-phospho-choline production.

Keywords: neuroprotection, cholest-4-ene-3,6-dione, anti-ROS, lipidomics, molecular docking

Introduction

Cerebral stroke is an acute cerebrovascular disease with the characteristics of high disability, mortality, and recurrence.¹ Clinically, approximately 75% of strokes are caused by focal cerebral ischemia due to arterial occlusion.² The current approaches for ischemic stroke mainly include restoration of blood flow, neuroprotection, and cell replacement therapies throughout the acute recovery progress,^{3,4} during which neuroprotection is considered an indispensable treatment.

The pathophysiological mechanisms of ischemic stroke mainly include energy depletion, nerve excitotoxicity, calcium overload, oxidative stress, blood-brain barrier disruption, inflammation, and nerve cell apoptosis,⁵ which have been targets for the development of neuroprotectant drugs. However, there are still few neuroprotectants that have been clinically translated. One of the possible reasons for the lack of clinical neuroprotectants is that a single-effect neuroprotectant fails to fully protect against an irreversible cascade of neuronal damage.⁶ Therefore, there is an urgent need to investigate novel comprehensive neuroprotectants for the treatment of ischemic stroke.



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