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Soluble N-ethylmaleimide-sensitive Factor Attachment Receptor (SNARE) Protein Involved in the Remission of Depression by Acupuncture in Rats

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Abstract

This study aims to investigate the molecular mechanisms of acupuncture in the remission of depression. A depressive disorder model was induced by exposing Sprague–Dawley rats to chronic unpredictable stress. The rats were divided into five groups: healthy (blank group) and stressed rats (model group), and stressed rats treated with acupuncture (acupuncture group), riluzole (riluzole group), acupuncture combined with botulinum toxin A (BTX-A) injection (acupuncture + BTX-A group) or riluzole combined with BTX-A injection (riluzole + BTX-A group). Behavioral analysis showed significant differences in sucrose consumption, weight, and horizontal or vertical movements between the model and both the riluzole and acupuncture groups. No obvious differences between the riluzole + BTX-A and acupuncture + BTX-A groups were found. Moreover, no significance differences in glutamate content in the hippocampus were found among the riluzole + BTX-A, acupuncture + BTX-A and model groups ($p > 0.05$). Western blots and reverse transcription polymerase chain reactions were employed to detect protein and mRNA expressions of VGLUT2, SNAP25, VAMP1, VAMP2, VAMP7, and syntaxin1; no obvious differences among the riluzole + BTX-A, acupuncture + BTX-A and model groups were found. These data suggest that soluble N-ethylmaleimide-sensitive factor attachment receptor proteins are involved in the remission of depression in rats treated with acupuncture.

1. Introduction

Depressive disorder is a mental disorder characterized by persistent low mood and is very difficult to cure. So far, the etiology of depression remains unclear. According to a World Health Organization report, the rate of global depression is about 11%, which makes it the world's fourth major disease [1]. Antidepressants, such as triple-reuptake inhibitors and riluzole, are the mainstay in the treatment of depressive disorders. However, the outcomes of treatment are unsatisfactory [2–4]. Therefore, alternative treatments for depressive disorders must be sought. Previous studies have found acupuncture to be an effective treatment modality for remitting stress disorders [5]. However, the mechanism underlying this effect remains ambiguous.

Extremely high levels of glutamate have been found in depressive patients, and clinical studies have indicated that depressive disorders are associated with increased glutamatergic activity [6]. Glutamatergic neurotransmission occurs predominantly within the confines of a tripartite synapse. It is then recycled through the glutamate–glutamine cycle by glutamate receptors in postsynaptic neurons. The interactions among synaptic soluble N-ethylmaleimide-sensitive factor attachment receptor (SNARE) proteins, synaptobrevin (vesicle-associated membrane protein, VAMP), synaptosomal associated protein 25 (SNAP-25), and syntaxin in the plasma membrane play important roles in glutamate neurotransmission. Dysregulation of the glutamatergic system has been found to be an important factor in depressive disorders [7].

Acupuncture is an ancient Chinese medical technic that can be traced back at least 2500 years. It is now widely used as a form of complementary and alternative medicine in many countries [8,9]. Clinical studies have found acupuncture to be an effective treatment modality for remitting stress disorder, anxiety, and depression [10–13]. Electroconvulsive therapy might regulate the expression

of distinct neurotrophic signaling pathways, including the metabotropic glutamate receptor, neuron glucose transporter, and Gamma aminobutyric acid A (GABA-A) receptor. Despite the effectiveness of acupuncture in the treatment of depressive disorders, very few studies have investigated the underlying mechanisms.

To probe into the mechanism of acupuncture in the treatment of depressive disorders, we investigated synaptic SNARE proteins and the glutamate–glutamine cycle. Botulinum toxin A (BTX-A), secreted by the deadly botulinum, is a highly toxic bacterial endotoxin. Evidence has shown that BTX-A is able to specifically hydrolyze SNARE proteins [14,15]. In this study, BTX-A was used to regulate the SNARE protein content and to investigate the molecular mechanisms of acupuncture.

2. Materials and methods

2.1. Animals

Adult, male Sprague–Dawley rats (180–220 g) were used in this experiment. Animals were obtained from the Experimental Animal Center of Guangzhou University of Chinese Medicine, Guangzhou, China. Both the animal's care and the study protocol were conducted according to the Helsinki Declaration Accords and followed the guidelines of the Committee on the Care and Use of Laboratory Animals at the Experimental Animal Center of Guangzhou University of Chinese Medicine. Animals were housed under a 12 hour light/dark cycle at a constant room temperature of 23°C.

2.2. Depressive disorder model and treatment

Depression disorder was induced in the rats by exposing them to chronic unpredictable stress (CUS) for 21 days [16]. The CUS included cold swims (5 minutes), water deprivation

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