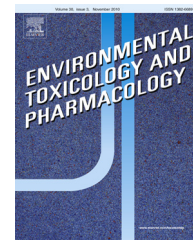


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Chronic unpredictable mild stress impairs erythrocyte immune function and changes T-lymphocyte subsets in a rat model of stress-induced depression

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ABSTRACT

Stress has been shown to suppress immune function and increase susceptibility to inflammatory and psychiatric diseases. This study sought to investigate the changes in erythrocyte immune functions and T-lymphocyte subsets and to explore the mechanism implicated in the process of stress-induced depression by employing a rat depression model induced by chronic unpredictable mild stress (CUMS). The body weights and behavioral changes of the rats were recorded, and plasma corticosterone levels were determined by radioimmunoassay. Erythrocyte immune function and T-lymphocytes subsets were respectively measured by the method of yeast rosette and flow cytometry at different time intervals, and their relationship was analyzed. Results indicated that a reduction was observed in the following: the rats' crossing and rearing movement times, the volume of sucrose intake and the preference for sucrose in the depression model group. Plasma corticosterone levels were elevated; the rate of E-C3bR decreased, and E-IC was increased. Some alterations in the percentage of T-lymphocytes and IL-2 appeared in the depression model group and some relationships existed between these parameters. Collectively, these findings disclose that long-time stress could induce changes in rat behavior and activities through an effect on erythrocyte immune functions and T-lymphocyte subsets.

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1. Introduction

Stressful experiences, particularly chronic and unintended stressors, are significant risk factors that play a pervasive role in the etiology of the myriad of diseases that they produce and exacerbate (Anon., 2014). Depression is the leading cause of disability and the 4th leading contributor to the global burden of disease in 2000 (Ren et al., 2008). Chronic stress is significantly associated with the development of mood and anxiety disorders and psychiatric disease (Anisman et al., 2008a). It has been reported that both major and minor depressive disorders raise the risk of cardiac disease-related mortality in patients with and without cardiac diseases at the baseline, and the excess mortality risk was more than twice as high for major depression versus minor depression (Wu et al., 2012). Chronic stress is associated with a neurobehavioral syndrome that is suggestive of depression and multiple processes ranging from psychic-related disorders to activation of the inflammatory immune system (Anisman et al., 2008b). Several studies had consistently indicated that psychological, behavioral and neurobiological profiles of depression were linked to the effects of inflammatory cytokines (Maes, 2009; Maes et al., 1997; Powrie et al., 1994). However, the pathophysiologic mechanism of depression pertaining to the interactions between the nervous and immune systems remains largely unknown. In the present study, a regimen of chronic unpredictable mild stress (CUMS), which was initially described by Willner et al. (1987) and termed chronic mild stress (CMS), was used as a reliable experimental model of depression (Willner, 1997; Willner et al., 1992). Most importantly, CUMS has been shown to mimic daily hassles and stress levels in humans.

In the early 1940s, special attention was drawn to the immune system with regard to physiological and psychological stresses. Strengthening the immune system will help prevent diseases (Irwin and Miller, 2007). The immune system, as the body's defense barrier, plays a crucial role in maintaining health, and is susceptible to adverse influences of the outside world. Research proved that the immune function could either enhance or weaken the mood. However, different animal species suffer from different kinds of stress, the immune function of the indicators is changing, and thus the results are not consistent (Katz et al., 1981). Acute stress can enhance the performance of the immune response whereas chronic stress, affects the immune system through changes in the neurohumoral system, and brings about immunosuppression (Denomme, 2004).

Erythrocytes are the most numerous blood cells in the blood and are required for tissue respiration (Greer et al., 2009). The natural immune function of erythrocytes can guide leukocyte natural immune response and acquired immune response (Guo et al., 2002). Erythrocyte immunity is an important part of nonspecific immunity in vertebrates and sensitive to environmental changes and drug exposure (Sun et al., 1994). Since the erythrocyte immune system was put forward, the relationship between the nervous system and erythrocyte immune function has become clearer (Guo and Chen, 2006; Hou, 2005; Deng et al., 2013), and the immune function is mainly achieved by the erythrocyte complement receptor type one (CR1) on the erythrocyte membrane (Reinagel and Taylor,

2000). Thus far, very limited studies have been conducted to examine the effects of chronic stress on the erythrocyte immune adherence function in stress. Nonetheless, further attempts should not only be confined to investigating the effects of chronic psychological stress on erythrocyte function in animals and humans, but should also explore the relationship between erythrocyte immune function and T-lymphocyte subsets in stress.

Therefore, the present study aimed to observe the behavioral changes and evaluate the effects on erythrocyte immune function, T-lymphocyte subsets and IL-2 in a chronic unpredictable mild stress rat model. Moreover, the mechanisms of the immune function in the process of stress-induced depression were also addressed.

2. Materials and methods

2.1. Animals

Twenty male Sprague-Dawley rats weighing 200 ± 20 g (supplied by the Animal Laboratory Center of Xinjiang Medical University) were randomly allocated, after acclimatization for a week, into 2 groups, namely a control group and a CUMS model group, with 10 rats in each group. Control rats were housed 5 per cage, while the model rats were housed individually. All rats were maintained under standard laboratory conditions (12 h light/dark cycle, temperature $21\text{--}23^\circ\text{C}$, relative humidity 45–65%, and food and water ad libitum except for model rats under deprived procedure).

2.2. Chronic unpredictable mild stress (CUMS) procedure

The CUMS procedure followed a previously described method (Willner et al., 1990) with minor modifications. Chronic unpredictable mild stress consisted of exposure to the following stressors in a random order everyday for 3 weeks: food deprivation for 24 h; water deprivation for 24 h; light and shade upside down for 24 h; noise housing (1500 Hz, 92 db, for 1 h); behavioral restriction for 4 h; forced swimming for 5 min in 4°C water; squeezing tail for 1 min. During the process, the model rats were moved into another room (light intensity and temperature of two room were basically the same), then back to the room after the stimulation.

2.3. Preparation of blood samples

Venous blood samples (2 ml) were collected from the rats on the day before stress (Baseline) and then on the 1st, 7th, 14th, and 21st day. The blood samples were divided into two aliquots: one was reserved for preparation of red blood cell suspension, and the other was used in the test of the lymphocyte subsets and IL-2. Plasma was used for determination of plasma corticosterone level.

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