Neuroscience 248 (2013) 252–260

EFFECT OF MATERNAL DEPRIVATION ON ACETYLCHOLINESTERASE ACTIVITY AND BEHAVIORAL CHANGES ON THE KETAMINE-INDUCED ANIMAL MODEL OF SCHIZOPHRENIA

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Abstract—Maternal deprivation has been associated with physiological and developmental changes that may be related to an increased risk for childhood and adult neuropsychiatric diseases. A growing number of studies demonstrated the importance of childhood experiences in the development of psychosis and schizophrenia in adulthood. Therefore, the present study investigated different behavior responses in rats following maternal deprivation and/or ketamine treatment in adulthood. Male rats were subjected to maternal deprivation for 180 min from postnatal day-01 to postnatal day-10. We evaluated locomotor activity, avoidance task and social interaction of adult male rats deprived or not deprived that were administered with saline or acute subanesthetic doses of ketamine (5, 15 and 25 mg/kg, i.p.). Our results show that only ketamine (25 mg/kg, i.p.) treatment in the adult rats lead to hyperlocomotion but not ketamine (5 and 15 mg/kg) and maternal deprivation alone. However, maternally deprived rats treated with ketamine (5 mg/kg) induced hyperlocomotion. Additionally, ketamine (25 mg/kg) and maternal deprivation alone induced cognitive deficit in the avoidance task. Rats deprived of and treated with ketamine (5, 15 and 25 mg/kg) also lead to memory deficit. Moreover, ketamine (25 mg/kg) and maternal deprivation alone increased latency to start social behavior. However, ketamine (5 mg/kg) and maternal deprivation lead to an increase of latency to start social behavior. Biochemistry data showed that all doses of ketamine and ketamine plus maternal deprivation increased the acetylcholinesterase (AChE) activity in the prefrontal cortex, hippocampus and striatum. The major doses of ketamine associated with maternal deprivation induced a major increase of AChE activity. Together, our results suggest that animals subjected to maternal deprivation had an increased risk for schizophrenia-like behavior and cholinergic alteration.

Key words: schizophrenia, maternal deprivation, ketamine, locomotor activity, memory, acetylcholinesterase.

INTRODUCTION

Among the severe psychiatric disorders, schizophrenia is considered one of the most morbid and debilitating (Moghaddam and Jackson, 2003), with an estimated prevalence of 1% in the world population (Abi-Dargham and Laruelle, 2005). The causes and frequency of comorbidities in schizophrenia are very poorly understood (Schultz et al., 2007). Typical symptoms of schizophrenia can be divided into positive, negative and cognitive (disorganized). Positive symptoms are related to an excess or distortion of features commonly seen in normal individuals, characterized by delusions and hallucinations, while the negative symptoms indicate decrease or loss of normal functions, such as the blunting of affect. The bizarre behavior, inappropriate affect and disorganization of thought can be considered disorganized symptoms (Bickel and Javitt, 2009; Bradford, 2009).

It is suggested that individuals who develop the disorder in adulthood, already present delayed neurodevelopment, with lower Intellectual Quotient (IQ) and lower school performance, when compared to other children (Jones et al., 1994). In the adult stage, attention and memory deficits are the main cognitive symptoms presented by patients (Kay et al., 1987; Abi-Dargham and Laruelle, 2005; Bickel and Javitt, 2009; Bradford, 2009).

The dopamine theory, such as the first proposal, suggests an association between the positive symptoms of schizophrenia and dopaminergic hyperactivity, given the increase in the occupancy of type 2 dopamine receptors (D2) by dopamine in the observed disorder (Carlsson and Lindqvist, 1963; Angrist et al., 1974; Bradford, 2009). After decades, schizophrenia research identified that drugs which have specific dopaminergic receptors as targets are not sufficient for the treatment of schizophrenia, since they are only effective in reducing the positive symptoms, but no efficacy against the negative symptoms and cognitive disorder (Moghaddam and Jackson, 2003).

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Abbreviations: AChE, acetylcholinesterase; AcSCh, acetylthiocholine iodide; ANOVA, analysis of variance; CNS, central nervous system; D2, dopamine receptors type 2; IQ, Intellectual Quotient; NMDA, N-methyl-D-aspartate receptors; UNESC, Universidade do Extremo Sul Catarinense.

0306-4522/13 $36.00
http://dx.doi.org/10.1016/j.neuroscience.2013.05.059

Another hypothesis is that glutamate, the main excitatory neurotransmitter in the central nervous system (CNS), is involved in various cognitive processes linked to memory and perception (Dingledine et al., 1999; Bradford, 2009). Evidence suggests that alterations in the activity of glutamate receptors, N-methyl-D-aspartate (NMDA) may be related to memory impairment present in the disorder (Newcomer, 1999; Bickel and Javitt, 2009).

Decreased glutamate levels found in the CNS of patients with schizophrenia are among the evidences supporting the involvement of this neurotransmitter in the pathophysiology of the disorder (Kim et al., 1980; Bradford, 2009). In this context, the neurotransmitter acetylcholine plays an important role in the cognitive processes (Gold, 2003; Prado et al., 2006; Benetti et al., 2009). Studies indicate the involvement of cholinergic mechanisms in learning and memory functions, also with a key role of both muscarinic and nicotinic receptors (Brown et al., 2002; Barros et al., 2005).

The involvement of the cholinergic system in the pathophysiology of mental disorders becomes even more relevant. As an example, changes in the nicotinic acetylcholine receptors are observed in schizophrenia. The cigarette use is significantly higher when compared to the general population, and this difference remained significant when comparing schizophrenic patients to other psychiatric patients (Mccreadile, 2002; Lherena et al., 2003; Aguilar et al., 2005). Two hypotheses suggest that smoking may both reduce the side effects of anti-psychotics and also improve the symptoms of schizophrenia, particularly negative, cognitive and/or depressive symptoms (Aguilar et al., 2005). Therefore, research with acetylcholinesterase (AChE) inhibitors represents a promising possibility to treat diseases with cognitive impairment, such as schizophrenia and Alzheimer's disease (Giacobini, 2004; Borlongan et al., 2005).

The variation between 13% and 17% (Cannon and Murray, 1998; Cardno et al., 1999) in liability to develop schizophrenia can be explained by the theory based on environmental factors, both biological and psychosocial. A growing number of studies demonstrate the importance of childhood experiences in the development of psychosis and schizophrenia in adulthood (Wicks et al., 2005). Most of these, focus on child maltreatment (Read et al., 2001), reports of problems in adulthood bonds (Berry et al., 2007) and loss of parents during childhood (Erlenmeyer-Kimling et al., 1991; Agid et al., 1999). In addition, a potential cause for possible diseases in adulthood is the separation from the mother during childhood (Bowlby, 1973, 1988). Based on this evidence it is believed that maternal deprivation can induce the development of psychiatric disorders in adulthood, such as, in most studies, depression (Lambás-Señas et al., 2009).

The animal model of schizophrenia used in this study was induced by ketamine, a dissociative anesthetic that acts as a noncompetitive antagonist of the NMDA glutamate receptor. This receptor is found especially in the hippocampus and cerebral cortex – critical areas to the processes of humans' memory and cognition (Morgan et al., 2004). This substance is able to reproduce in humans and animals, which are associated with the symptoms observed in connection with schizophrenia (Park and Holzman, 1992; Newcomer, 1999; Becker et al., 2003; Farber, 2003). Behavioral changes induced by ketamine in rats that are related to symptoms and neurobiological markers of schizophrenia, include four main dimensions: hyperlocomotion, stereotypy, impaired information processing with cognitive functions of memory and attention, and impaired social interaction (Lipska and Weinberber, 2000; Bubeníková-Valesová et al., 2010).

Finally, the objective of this research topic becomes relevant in the importance of contributing to the neuroscience research, not the resolution of the problem in its full context, but in the search for drugs with less potential for weaknesses, effective psychotherapies in order to improve the quality of life and forms that enable greater integration of social interaction in schizophrenia.

EXPERIMENTAL PROCEDURES

Experimental procedures involving animals were performed in accordance with the NIH Guide for the Care and Usage of Laboratory Animals and recommendations of the Brazilian Society for Neuroscience and Behavior (SBNeC). Furthermore, the execution of the work was previously approved by the local Ethics Committee in usage of the Universidade do Extremo Sul Catarinense (UNESC).

Animals

Pregnant female Wistar rats (age of 2 months, weight of 200–220 g) were obtained from the breeding colony of UNESC, Criciúma, SC, Brazil. The dams were individually housed with sawdust bedding and *ad libitum* access to food and water. After the birth of the litter, the offspring was separated. Only male offspring were included in the study.

Animal model of schizophrenia

Symptoms of schizophrenia were induced by subanesthetic doses of ketamine. Acute ketamine (5, 15 and 25 mg/kg doses, i.p., CU Chemie Uetikon, Germany) was administered at a volume of 1 mL/100 kg in male adult offspring. Ketamine (25 mg/kg) induces symptoms of schizophrenia as hyperlocomotion, social isolation, and cognitive deficit, demonstrated in previous studies (Hunt et al., 2006; Imre et al., 2006; de Oliveira et al., 2011).

Maternal deprivation

At the first postnatal day, after withdrawal of female offspring, the male offspring was separated into two groups: (1) deprived and (2) non-deprived. The first group was deprived of the mother for 3 h a day for the