Brain Research Bulletin 110 (2015) 40–46

Contents lists available at ScienceDirect

Brain Research Bulletin

journal homepage: www.elsevier.com/locate/brainresbull

Research report

Lateral habenula as a link between dopaminergic and serotonergic systems contributes to depressive symptoms in Parkinson's disease

Xiao Feng Luo [1], Bei Lin Zhang [1], Ji Cheng Li, Ying Ying Yang, Yan Fei Sun, Hua Zhao *

Department of Physiology, Key Laboratory of Pathobiology, Ministry of Education, College of Basic Medical Sciences, Jilin University, Changchun 130021, PR China

ARTICLE INFO

Article history:
Received 28 August 2014
Received in revised form
26 November 2014
Accepted 29 November 2014
Available online 8 December 2014

Keywords:
Lateral habenula
6-hydropamine rat model
Depressive-like behavior
Raphe nuclei
Substantia nigra
Serotonin

ABSTRACT

Degeneration of substantia nigra dopaminergic neurons is a key pathological change of Parkinson's disease (PD), and its motor consequences have been widely recognized. Recently, mood disorders associated with PD have begun to attract a great deal of interest, however, their pathogenesis remains unclear. PD is associated with not only degenerative changes in dopaminergic neurons in the substantia nigra but also changes in serotonergic neurons in the raphe nuclei. The abnormalities in central 5-hydroxytryptamine (5-HT) neurotransmission are thought to play a key role in the pathogenesis of depression. The lateral habenula (LHb) is closely related to the substantia nigra and raphe nuclei, and its hyperactivity is closely related to the pathogenesis of depression. In this study, we screened rats with depressive-like behaviors from PD model animals and found that cytochrome c oxidase activity in the LHb of these rats was twice that seen in the control rats. In the forced swim test, LHb lesions caused a decrease in depressive-like behavior of PD rats as indexed by decreased immobility times and increased climbing times. Additionally, LHb lesions caused an enhance in 5-HT levels in the raphe nuclei. These results suggest that LHb lesions may improve depressive-like behavior in PD rats by increasing 5-HT levels in the raphe nuclei. Thus, LHb contributes to the depressive-like behavior in PD rats via mediating the effects of dopaminergic neurons in the substantia nigra on serotonergic neurons in the raphe nuclei.

1. Introduction

Parkinson's disease (PD) is a degenerative disease of the central nervous system that is common in the elderly population. The motor symptoms of PD are well recognized and have been the focus of much research. However, the mood disorders observed in most PD patients, such as depression (Shulman et al., 2001; McDonald et al., 2003), should not be neglected because they seriously affect the quality of life and prognosis of PD patients; furthermore, they can be a direct cause of death in these patients (Carod-Artal et al., 2007; Rahman et al., 2008).

Numerous studies have shown that movement disorders result from reductions in dopamine levels in the striatum, which are caused by degenerative pathological changes in midbrain substantia nigra pars compacta dopaminergic neurons. Therefore, the clinical application of levodopa can obviously improve movement disorders in PD patients (Zibetti et al., 2013). However, PD-related affective disorders are associated with not only disrupted dopamine systems but also nondopamine system changes, such as central 5-hydroxytryptamine (5-HT) system disorders (Scatton et al., 1983). It is widely known that a decreased 5-HT level in the raphe nuclei is related to the onset of depression (Stockmeier et al., 1998; Gray et al., 2013). Interestingly, Mayeux et al. (1984) have shown that 5-hydroxyindoleacetic acid levels in the cerebrospinal fluid of PD patients with depression were lower than those in the normal controls and nondepressed PD patients. In addition, serotonin reuptake inhibitors can markedly improve depressive-like symptoms in PD patients (Devos et al., 2008). Consequently, depression symptoms in PD patients may be the result of the interaction of the two systems. Recently, Bastide et al. (2014) have reported that immediate early gene levels, such as ΔFosB, change not only in basal ganglia structures but also in other brain areas of PD-model animals given levodopa to induce hyperactive behavior,

Abbreviations: PD, Parkinson's disease; PDD, Parkinson's disease with depressive-like behavior; LHb, The lateral habenula; SN, substantia nigra; HRP, horseradish peroxidase; FST, forced swim test; OFT, open-field test; CCO, cytochrome c oxidase; MFB, medial forebrain bundle; 5-HT, 5-hydroxytryptamine; RN, raphe nuclei.

* Corresponding author at: Department of Physiology, College of Basic Medical Sciences, Jilin University, 126 Xinmin Street, Changchun 130021, PR China. Tel.: +86 431 85619472.
E-mail address: zhua@jlu.edu.cn (H. Zhao).

[1] These authors contributed equally to this work.

http://dx.doi.org/10.1016/j.brainresbull.2014.11.006
0361-9230/

including the lateral habenula (LHb). The structure may link the substantia nigra dopamine system with the raphe nucleus serotonin system, thus playing important roles in PD patients with depressive-like behavior (PDD).

The LHb directly projects into the substantia nigra and raphe nucleus and it is involved in a wide range of functions related to the two structures, including cognitive and emotional functions, pain sensitivity and sleep and circadian rhythm regulation (Lecourtier et al., 2004; Shelton et al., 2012; Zhao and Rusak, 2005; Aizawa et al., 2013). In recent years, increasing attention has been paid to the role of the LHb in depression (Yang et al., 2008). It has been reported that LHb activity is enhanced in animals with depressive-like behaviors as well as in depressed patients (Caldecott-Hazard et al., 1988; Morris et al., 1999). Moreover, high-frequency deep-brain stimulation of the habenula to inhibit its activity was able to alleviate depressive symptoms in patients who could not be improved by antidepressants (Sartorius et al., 2010). Additionally, in animal models, LHb lesions were shown to improve depressive-like behavior by increasing raphe 5-HT levels (Yang et al., 2008).

To date, the relationships between the LHb and substantia nigra dopaminergic neurons, especially in reward processing, have been an area of focus as these two systems inhibit each other and act in distinct ways (Matsumoto and Hikosaka, 2007). Lesions of the substantia nigra (SN) have been shown to increase LHb glucose use rates, whereas efferent signals from the medial globus pallidus to the LHb in 6-hydroxydopamine (6-OHDA)-induced PD animals were found to be increased (Wooten and Collins, 1981). Therefore, the LHb not only directly controls substantia-nigra and raphe-nuclei activities, but it also acts as a bridge for communication between the substantia nigra dopamine system and the raphe nuclei serotonin system. Accordingly, substantia nigra dopaminergic neuron losses in PD rats were hypothesized to reverse its inhibitory effect on LHb, thereby increasing LHb suppression on the raphe neurons and decreasing 5-HT levels in the raphe nuclei. In this study, the effects of LHb lesions on depressive-like behavior and the 5-HT concentration in the raphe nuclei in PD rats were observed to determine whether the hyperactivity of LHb is involved in the pathogenesis of depression associated with PD by enhancing its inhibition on raphe nuclei 5-HT neurons.

2. Materials and methods

2.1. Animals

Male Wistar rats (Department of Experimental Animals, Jilin University, Changchun, China) were used in this study. Rats were housed in a standard cage, with free access to food and water, in a temperature-controlled room ($23 \pm 1\,^{\circ}C$) on a 12-h light/dark cycle (lights on at 8:00 am).

A total of 130 Wistar rats, 9–10 weeks of age and weighing 200–230 g, were used in this study. Of all these rats injected with 6-OHDA into brains, two died before the rotation test (a mortality rate of 1.5%). We obtained 61 PD rats from 128 rats injected with 6-OHDA and screened 26 rats with depression-like behavior from 61 PD rats by using the forced swimming test. Animals were randomly divided into four experimental groups: the control group (control, $n = 10$), PD with depressive-like behavior group (PDD, $n = 7$), PDD and electrical bilateral lesions of the LHb group (PDD + LHb lesion, $n = 13$), and PDD and sham lesions of the LHb group (PDD + LHb sham, $n = 6$).

All procedures used in this study were conducted in accordance with the international standards for animal welfare and were approved by the local Committee for Animal Care Research at Jilin University. All efforts were made to minimize the number of animals used and their suffering.

Apomorphine and 6-OHDA were obtained from Sigma (St. Louis, MO, USA).

2.2. Animal model of PD

Animals were anesthetized with sodium pentobarbital (60 mg/kg, intraperitoneally). To protect noradrenergic neurons, rats were pretreated with desipramine (25 mg/kg, intraperitoneally) 30 min before 6-OHDA was injected. The pretreated rats were fixed in a stereotaxic instrument, then the scalp was incised to expose the skull, and a single hole was drilled over the right medial forebrain bundle (MFB) (4.0 mm posterior to the bregma; 1.65 mm lateral to the midline; 8.0 mm ventral to the dura surface; Paxinos and Watson, 1998). Four microliters of 6-OHDA hydrochloride solution (4 μg/μL) containing 0.2% ascorbic acid was injected into the right MFB with a microinjector. The microinjector was left in place for an additional 8 min before being slowly withdrawn.

After the operation, the rats were monitored until they reached consciousness and then they were returned to their cages. Other than only being injected with the solvent, the sham-operated rats were treated in an identical manner to the model rats. The rats were submitted to a rotational test induced by apomorphine (0.1 mg/kg, intraperitoneally) at the end of the 4th week after the 6-OHDA injection. Only the rats that exhibited a contralateral rotation rate greater than 7/min were considered to have hemiparkinsonism, and these rats were selected for further study.

2.3. Behavioral experiments

In this study, we used the forced swim test (FST) and open-field test (OFT) to assess depressive-like behavior in the PD model. The animals in the control and the PDD groups were submitted only once to the behavioral tests, and the other two groups (PDD + LHb lesion and PDD + LHb sham) were submitted twice to the behavioral tests. We did not find a significant difference between twice behavioral tests, which is consistent with the results in our previous experiments (Yang et al., 2008) and also the results reported by Armario et al. (1988). Since we had an additional PDD + LHb sham group as control, the repeated tests should not have affected the behavioral results.

2.4. Forced swim test

The rats were placed in a clear cylinder (20-cm internal diameter, 50 cm high) filled with $24 \pm 1\,^{\circ}C$ water to a depth of 30 cm, according to a previous report (Porsolt et al., 1978). To avoid interference from feces and other floating objects, the water was changed after each experiment. A 15-min pretest followed by a 5-min test was conducted the next day. Only the times spent immobile and climbing during the 5-min test were measured to determine the behavioral score. A digital video camera was mounted above the cylinder, and recorded files were used for future analyses. After the pretest and test sessions, the animals were taken out of the water, dried with a paper towel, and returned to their cages.

2.5. Open-field test

According to a previous report (Katz et al., 1981), the animals were placed in the center of an open-field chamber (100 cm × 100 cm × 50 cm) with the inside walls painted black. The chamber floor was divided by black lines into 25 equal squares to assess locomotion. Each rat was tested over 3 min. The movements of the rats in the open field were quantified by counting the number of crossings (all four paws in a square) and the number of rearings