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Neurotransmitter changes and behavioral manifestations in mouse models of Parkinson's disease

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Abstract: This paper studies the association of neurotransmitter changes with behavioral manifestations on a mouse model for Parkinson's disease. The experimental results showed mice in the Parkinson's disease model group had significantly reduced dopamine content and decreased motor functions. Dopamine content correlated positively with total moving distance and rotarod holding time. These results suggest a direct link between decline in motor function and damage to dopaminergic neurons. The results carry out the close relationship between damage in dopaminergic neurons and movement disorder during Parkinson's disease and offer essential experimental bases for the treatment of the same disease. The present research will, hence, be of theoretical value in the basis for Parkinson's disease studies by revealing the close relationship between neurotransmitter changes and motor dysfunction in Parkinson's disease and indicating directions to follow in further investigations.

1. Introduction

1.1 Research background and importance

Parkinson's disease (PD) is a neurodegenerative disorder, characterized by dopaminergic neuron loss and related symptomatology [1]. Although the exact cause remains elusive, degeneration of these neurons and relevant neurotransmitter changes are key in PD's pathology. Mouse models have been instrumental in studying PD's mechanisms and potential treatments, simulating it via chemical induction of toxins like MPTP or 6-OHDA [2]. The MPTP mouse model, in particular, has become popular for its ease of use and similar pathological features to human PD [3-4]. Many studies have revealed that the levels of monoamine neurotransmitters, such as dopamine, norepinephrine, and serotonin, alter dramatically in PD mouse models; these changes are related directly to behavioral performance [5]. For example, with declining motor functions, a significant reduction in dopamine levels is seen, while the changes in norepinephrine and serotonin are associated with non-motor symptoms such as depression and anxiety [6]. Using these models, the pathology mechanisms of PD can be understood in detail, and we are able to screen the efficacy and safety of new drugs.

1.2 Research objectives

This research would seek to establish the systematic probing of the relationship between

neurotransmitter changes to behavioral manifestations in a mouse model of Parkinson's disease. More precisely: measuring the content of dopamine in mice in the Parkinson's diseases mouse model group; assessing its motor function; and elucidating the relationship between these two regarding motor behavior indicators, such as total moving distance and rotarod holding time, using correlation analysis. This study's findings on the link between dopamine neuron damage and motor dysfunction offer valuable insights for Parkinson's disease treatment and further research.

2. Literature Review

2.1 Pathogenesis of Parkinson's disease

Parkinson's disease (PD) has a complex and varied pathogenesis. It mainly involves loss of dopaminergic neurons in the substantia nigra pars compacta, forming Lewy bodies from aggregated α-synuclein [7-8]. These changes link to mitochondrial dysfunction, protein clearance defects, and neuroinflammation. Mitochondrial dysfunction is a causative factor in PD, leading to neuronal degeneration through decreased bioenergetic function, impaired quality control, and endoplasmic reticulum-mitochondrial process disorder. Intestinal inflammation also relates closely to PD pathogenesis via the gut-brain axis mechanism [9-10]. PD represents a combination of gene actions and environmental factors, including toxin exposure and mutations that induce or promote the diseases. The pathogenesis reflects a complex process of multiple factors and interactions, warranting further research into specific molecular and cellular mechanisms [11].

2.2 Research progress on mouse models of Parkinson's disease

The mouse model for understanding Parkinson's disease is essential, primarily in simulating the pathological characteristics of Parkinson's in humans and experimenting drug therapies [12-13]. One commonly used model is the 1-methyl-4-phenyl-1, 2, 3, 6-tetrahydropyridine (MPTP) mouse model. With this model, we can investigate selective nigrostriatal dopaminergic cell death, either induced systemically or locally, and it produces the movement disorder syndrome seen in Parkinson's patients. Indeed, the injection of different doses and regimens of MPTP treatment has been found to produce significant changes in the motor behavior performance and neurochemical indicators of mice [14]. Moreover, looking at their immense application in studying genetic mechanisms as well as mitochondrial dysfunction in relation to PD, indeed genetically modified mouse models do apply even further, including PINK1 mutant and Parkin mutant mouse models [15]. These models are therefore quite significant in verifying genes related to Parkinson's disease and screening potential therapeutic drugs. Along this line of thinking, in recent years researchers have also attempted to simulate the multi-causal characteristics of PD through the construction of multi-factor models, for example, the generation of a genetic mouse model in combination with MPTP treatment, in an effort to more fully reflect the underlying pathological mechanism of the disease [16].

2.3 Changes in neurotransmitters in Parkinson's disease

Neurotransmitter change is one of the important areas for research in characterizing the pathological process of Parkinson's disease. It has been reported that there exists a remarkable decline in nigrostriatal dopaminergic neuron in patients with PD, which finally leads to a sharp decline in the striatal dopamine level and is considered to be the main cause of motor symptoms [17]. Besides, other monoamine neurotransmitters, like norepinephrine and serotonin, also change obviously in PD, and these changes were closely related with non-motor symptoms such as depression and anxiety [18]. Decreases in these levels of dopamine are commonly associated with decreased motor functions in

behavior tests, such as in mouse models showing worsened performance in the rotarod and open field tests. Additionally, the level of dopamine metabolites, like DOPAC and HVA, changed drastically in Parkinson's disease mice models, indicating a disturbance in the dopamine metabolic pathway [19]. Quantitative analysis of neurotransmitters through techniques such as high-performance liquid chromatography may be required in order to probe more deeply into the pathologic mechanisms and the likely therapeutical targets of Parkinson's disease [20].

3. Experiment

3.1 Experimental materials and methods

3.1.1 Experimental animals and their treatment

The study used C57BL/6J mice, weighing 20-25g, and randomly divided them into a control group and a Parkinson's disease model group. The Parkinson's group was established by intraperitoneal injection of 1-methyl-4-phenyl-1, 2, 3, 6-tetrahydropyridine (MPTP). The control group received saline injections.

3.1.2 Experimental reagents and equipment

The main reagents used in the experiment include: 1xPBS, PFA, sucrose, urethane, OCT, DAPI, PBST, goat serum, ethanol, isopropanol, nucleic acid dye, HCL, solid formaldehyde, agarose. The main equipment includes: dissecting scissors, ophthalmic forceps, pointed forceps, scalpel, 24-well plate, slide, cover glass, disposable syringe, tin foil, centrifuge tube, measuring cylinder, measuring cup, electrophoresis instrument, plastic dropper, pipette, frozen slicer, fluorescence microscope, VS200, PCR instrument.

3.1.3 Mouse genotype identification

Genotype identification is performed by cutting the mouse tail. After lysing with lysis buffer for 2-3 hours, add isopropanol and shake centrifuge. After centrifugation, discard the supernatant, add ethanol and centrifuge again, and then dry. Amplify the mouse genotype by PCR, and run the gel to detect the amplification results.

3.2 Experimental steps

3.2.1 Mouse perfusion and brain tissue sampling

The steps of heart perfusion are as follows:

① Anesthetize the mouse and confirm that it is in a deep anesthesia state.

② Fix the mouse on the foam table and expose the heart.

③ Perfuse with saline for 10 minutes to drain the blood, and then perfuse with 4% PFA for 40 minutes to fix the tissue.

④After the perfusion, remove the mouse brain tissue and fix it with PFA.

3.2.2 Cryosectioning and DAPI staining

① Dehydrate the fixed brain tissue and place it in 30% sucrose solution.

② Embed the brain tissue in OCT and place it in a cryostat for sectioning (40 microns).

③ Stain the sections in DAPI solution for 15 minutes and then wash with PBS.

3.2.3 Immunofluorescence staining

① Place the brain slices in a blocking solution (a mixture of goat serum and PBST) and block for 2 hours.

② Incubate with primary antibody (NeuN antibody, rabbit source) overnight, then wash with PBST.

③ Incubate with secondary antibody (GAR-488, 1:500) for 2 hours, then wash with PBST.

④ Perform DAPI staining again, and finally wash with PBS for 10 minutes.

⑤ Spread and seal the slices, wash with PBS, add sealing agent and cover with coverslip.

4. Results and Analysis

4.1 Analysis of neurotransmitter changes

Dopamine content analysis

The dopamine content in brain tissue of mice in a control group and a Parkinson's disease model group was determined by high-performance liquid chromatography (HPLC). The results (Table 1) showed the dopamine content of mice in the Parkinson's disease model group was significantly reduced.

Table 1: Comparison of dopamine content in the control group and Parkinson's disease model group

Group	Dopamine Content (ng/mg)
Control	150 ±10
Parkinson's	60 ±5

Comparison of dopamine content in brain tissue of mice in the control group and Parkinson's disease model group. The dopamine content of mice in the Parkinson's disease model group was significantly lower than that of the control group ($p < 0.01$).

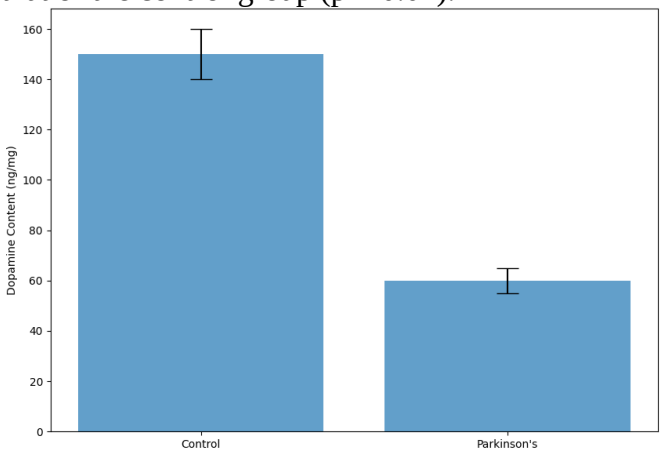

Figure 1: Changes in dopamine content

As shown in Figure 1, the brain tissue of mice in a Parkinson's disease model group showed significantly reduced dopamine content compared to the control group, reflecting damage to dopaminergic neurons.

4.2 Behavioral performance analysis

Open field test results

Open field test results showed that the total moving distance of mice in the Parkinson's disease model group was significantly reduced, indicating that their movement ability was impaired.

Table 2: Open field test results

Group	Total Distance (cm)
Control	500 ± 50
Parkinson's	300 ± 40

As shown in Table 2, the total moving distance of mice in the Parkinson's disease model group was significantly lower than that in the control group in the open field test, indicating reduced exploratory and motor activity. The rotarod experiment results in Table 3 showed the retention time of mice in the Parkinson's disease model group on the rotarod was significantly shortened, further demonstrating reduced motor coordination ability.

Table 3: Rotating rod experimental results

Group	Time on Rotarod (s)
Control	120 ± 10
Parkinson's	80 ± 15

Table 3 shows the retention time of mice in the control group and the Parkinson's disease model group during the rotarod experiment. The retention time of mice in the Parkinson's disease model group was significantly lower than that in the control group ($p < 0.01$).

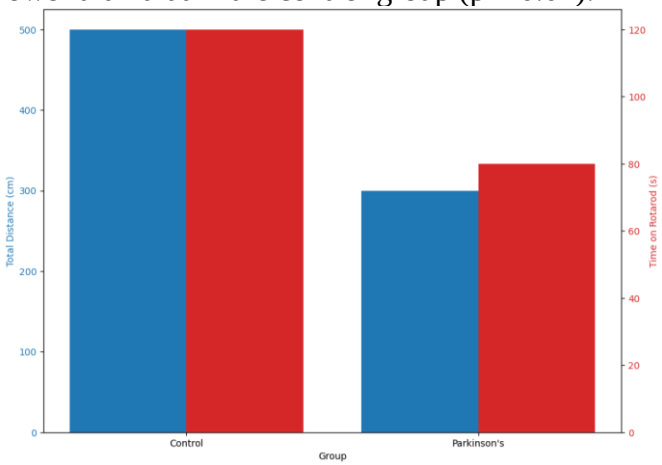

Figure 2: Behavioral test result

Figure 2 shows behavioral test results. Blue bars represent total distance traveled in the open field test; red bars represent holding time in the rotarod experiment. Mice in the Parkinson's disease model group declined significantly in both tests.

4.3 Correlation analysis between neurotransmitter changes and behavioral performance

With a view to further understand how neurotransmitter alteration is related to behavioral performance, the Pearson correlation coefficient was first of all applied for analyzing the relations between neurotransmitter alterations and behavioral performance.

We quantified the correlation between changes of neurotransmitters and behavioral performances by using the Pearson correlation coefficient.

$$r = \frac{n(\Sigma xy) - (\Sigma x)(\Sigma y)}{\sqrt{[n\Sigma x^2 - (\Sigma x)^2][n\Sigma y^2 - (\Sigma y)^2]}} \quad (1)$$

Among them, r is the correlation coefficient, n is the sample size, x and y represent the data values of the two sets of variables respectively.

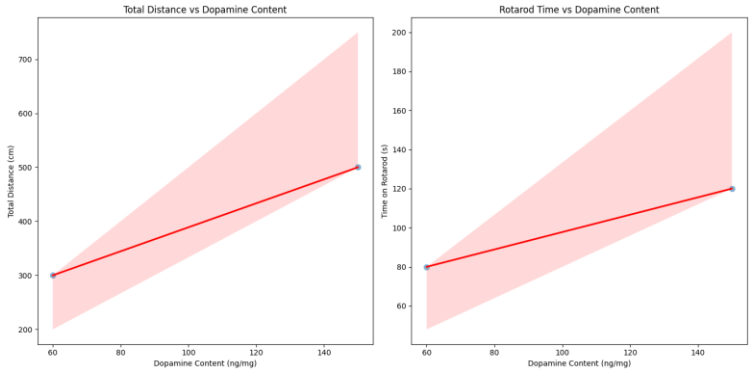

Figure 3: Correlation analysis between neurotransmitter changes and behavioral performance.

Figure 3 is a correlation analysis between neurotransmitter changes and behavioral performance. The left figure shows the positive correlation between dopamine content and total movement distance, and the right figure shows the positive correlation between dopamine content and rotarod retention time. The figure contains linear regression lines and scattered data, which further enhances the intuitiveness of the data and the reliability of the analysis. Both show that the higher the dopamine content, the better the motor function of the mouse.

5. Conclusion

This study aimed to understand and systematize changes in neurotransmitters with behavioral manifestations in a Parkinson's disease mouse model. Experimental results showed that, compared to the normal control group, the dopamine content in mice in the Parkinson's disease model group was significantly reduced. This suggests successful induction of the Parkinson's disease model of damage to dopaminergic neurons. Behavioral tests showed that, compared to the control group, the total moving distance of mice in the Parkinson's disease model group in the open field test was significantly lower, and the retention time in the rotarod test was significantly shortened, suggesting a severe decline in motor function after dopaminergic neuron injury.

Through correlation analysis, we further revealed the positive relationship between dopamine content and behavioral performance. The finding indicates neurotransmitter changes directly affect motor function in Parkinson's disease mice models. Higher dopamine levels correlated with greater athleticism. The result verifies the link between dopaminergic neuron damage and movement disorders in Parkinson's disease. It also provides important data for future treatment. Generally speaking, through this study, I representatively expose in detail the relativity between neurotransmitter change and Parkinson's disease-related motor dysfunction and fill out a theoretical basis of research into it individually.

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