

CONNECTING THE DOTS: MECHANISTIC INSIGHTS INTO EXERCISE TRAINING FOR POST-STROKE DEPRESSION IN RAT MODELS

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Abstract:

Depression and stroke are two leading contributors to the global socioeconomic burden. Depression, as per the World Health Organization, stands as the "foremost cause of disability worldwide," while stroke ranks among the top three contributors to the overall disease burden. Post-stroke depression (PSD) emerges as the most prevalent neurological and psychiatric complication following a stroke, presenting a spectrum of distressing symptoms, from apathy to self-harm, and even suicide. A recent cross-sectional meta-analysis, incorporating diverse geographical locations, evaluation time frames, and assessment methodologies, revealed a PSD prevalence ranging from 18% to 33%. Furthermore, a longitudinal meta-analysis uncovered that 55% of stroke survivors experienced depression at some point following cerebrovascular incidents, highlighting the cumulative incidence of PSD. Patients afflicted by post-stroke depression face formidable challenges, including elevated mortality rates, substantial cognitive impairment, increased disability risk, diminished quality of life, and heightened susceptibility to suicidal tendencies. The evidence underscores the urgency of addressing PSD to alleviate the physical, emotional, and societal burdens that it inflicts on stroke survivors.

Keywords: Post-stroke depression (PSD), Stroke, Depression, Cumulative incidence, Quality of life

Introduction

Depression and stroke are two important causes of socio-economic burden. According to the recent report of the World Health Organization, depression "is the main cause of disability worldwide" [1], and stroke is one of the three major causes of the Disease burden [2-3]. Post stroke depression (PSD) is the most common neurological and psychiatric complication after stroke [4-7], with the main symptoms being apathy, loss of appetite, drowsiness, self blame, self harm, and even suicide [8-9]. A recent cross-sectional meta-analysis based on different locations [10], evaluation time points [11-13], and evaluation methods estimated the prevalence of PSD to be between 18% and 33% [11-14]. Another longitudinal meta-analysis found that 55% of stroke patients had depression at a certain time point after cerebrovascular events (i.e., the Cumulative incidence of PSD was 55%) [11]. Compared with post stroke patients without depression, patients with post stroke depression have higher mortality rates (all-cause mortality rates of post stroke depression between 1.6 and 1.9) [13,15], more significant cognitive impairment [16], higher relative risk of disability, approximately 2.2 [17], lower quality of life [18], and higher suicidal tendencies [19]. Although systematic research on PSD has been ongoing for decades, many research results are still controversial due to the complex and diverse pathogenesis of PSD. A recent study suggests that the gut microbiota primarily regulates brain function and human behavior through a bidirectional communication pathway called the brain gut axis. These findings contribute to understanding mental disorders, especially depression [20]. At present, exercise training is the main means for functional recovery of stroke. Its mechanism is to cause correct functional reorganization of the brain under the exercise training guided by work through the characteristics of central Neuroplasticity changes and functional reorganization, including the recovery of motor function [21], depression [22], etc. Although there is no doubt about the effect of exercise training on PSD model recovery, whether its mechanism is related to Gut

microbiota is still unclear. Our research is based on the possible mechanism of Gut microbiota pottery altar exercise training to improve post-stroke depression.

1. Materials and Methods

1.1 Experimental animals

SPF grade rats were selected, with initial body weights ranging from 220g to 250g and daily age ranging from 45 to 55 days. Feeding conditions: The feeding room is well ventilated, with alternating light and dark for 12 hours, air humidity of 55% -65%, and temperature of 23-25 °C. All rats need to be fed adaptively for one week before the start of the experiment, and run on the treadmill for 3 days (speed 15m/min, time 30min/d). Screening can complete the treadmill training Rats practicing the plan.

1.2 Experimental grouping

60 rats were divided into blank group, Sham surgery group, post-stroke depression group, post-stroke depression+exercise training group by using random number table. There were 15 rats in each group. If the model fails or dies in the experiment, additional rats need to be selected and added to the group.

1.3 Model Preparation

1.3.1 Preparation of MCAO rat model

Strictly control the weight of rats within the range of 250-280g before modeling. Prepare a rat model of middle cerebral artery occlusion (MCAO) based on previous research experience. The mNss method was used 24 hours after surgery to assess the neurological deficits in MCAO rats. The ischemia-reperfusion experimental method (I/R) rat model was successfully created and placed in the same cage for 1 week, with normal diet and drinking water. For rats that died, were infected, or did not meet the scoring criteria during and after surgery and were excluded, the experimental quantity will continue to be supplemented.

1.3.2 Preparation of PSD rat model

The method for establishing depression models in I/R rats was improved based on literature, using chronic mild unpredictable stress (CUMS) combined with solitary confinement (single cage feeding) to establish a chronic stress PSD model. The stimulation methods include: (1) fasting; (2) Prohibition of water; (3) Tilt the mouse cage; (4) Continuous lighting; (5) Wet environment; (6) Ice water swimming; (7) Space limitations; (8) Pinch tail; (9) Noise stimulation; (10) Horizontal oscillation.

1.4 Intervention methods

Control group: No processing will be performed, only equal conditions will be given for grabbing. Sham group, only the skin was cut locally, and the incision was directly sutured after the carotid artery was free. After the operation, the animal was fed back without any treatment, and only the same conditions were given.

Model group: MCAO postoperative cage feeding+CUMS+solitary feeding, without any treatment. Intervention group: MCAO postoperative cage feeding+CUMS+solitary feeding. One day after successful modeling, electric treadmill training was used for exercise training, once a day, for 14 consecutive days. Slope: 0°; Speed: 8m/min in 1-3 days, 12m/min in 4-7 days, 15m/min in 7 days; Time: 30min/time.

1.5 Outcome

Sugar preference tests (SPT), forced swimming test (FST), and ELISA were used to detect the content of IL-2 and IL-6 in the peripheral serum of rats in each group, and high-throughput 16SrDNA Miseq sequencing analysis and QIIME software Recognition sequence were used to draw the composition information of Gut microbiota of rats in each group.

1.6 Statistical Analysis

Using statistical software (Graphpad prism7.0, Graphpad software, USA) to analyze data and measure. The data are expressed in terms of mean $\pm$ standard. The Levene's method is used for homogeneity of variance testing (if the variance is not homogeneous, rank sum test analysis is used). Two sets of data were compared using student's t-test; Multiple group comparisons were analyzed using one-way ANOVA and Tukey post hoc test. The significance level is set to $p < 0.05$.

2. Results**2.1 Behavioral status of rats in each group**

Table 1: Behavioral status of rats in each group

Groups	n	SPT(%)	FST(S)
Control	15	90.15±2.11	53.34±4.23
Sham	15	89.45±3.51	52.94±4.15
Model	15	49.19±1.24#	111.9±3.74*
Intervention	15	70.94±3.10#	79.56±3.21*
t		4.57	6.23
p		<0.001	<0.001

This study used SPT to assess lack of pleasure and FST to assess depression and despair towards CUMS after stroke. SPT and FST were better in the intervention group than in the model group, $p < 0.001$, as shown in Table 1.

2.2 Expression of inflammatory factors in different tissues of experimental animals

Table 2: Expression of inflammatory factors in different tissues of experimental animals

Groups	n	IL-1(pg/mL)		IL-6(pg/mL)	
		Intestina tissue	brain tissue	Intestinal tissue	brain tissue
Control	15	13.12±2.31	9.59±1.11	12.34±3.23	6.16±1.26
Sham	15	14.05±2.51	10.01±1.25	13.14±2.15	6.42±1.12
Model	15	78.19±7.24#	80.11±9.14^	6.91±4.71*	1.39±0.28*
Intervention	15	53.91±3.11#	57.22±6.71^	22.56±3.36*	11.76±2.26
t		7.99	4.17	8.68	9.20
p		<0.001	<0.001	<0.001	<0.001

The intervention group significantly inhibited the expression of inflammatory factors and upregulated the expression of anti-inflammatory factors in both intestinal and brain tissues, $p < 0.001$, as shown in Table 2.

2.3 Composition information of Gut microbiota

Compared with the model group, the expression levels of Lactobacillus and Bifidobacterium in the intervention group increased after exercise training in rats.

3. Discussion

Stroke is the main cause of adult disability in many countries [23], and it is second only to cancer and heart disease. Three major causes of death. Among them, ischemic stroke accounts for over 80% of stroke patients. Depression is the most common complication of ischemic stroke, with approximately one-third of people affected by post stroke depression (PSD) [24], characterized by a lack of pleasure, low self-esteem, fatigue, and a sense of worthlessness [25]. This has had a serious negative impact on the functional recovery and quality of life of survivors, increasing the suicide rate and disability rate, and seriously increasing the burden on families and society. The complexity of PSD mechanisms makes its prevention and treatment a challenging task. The conventional treatment of depression is selective serotonin Reuptake inhibitor (SSRIs). Although the mechanism of action is still controversial, it has been proved to be effective in preventing and treating PSD in clinical practice [26]. However, SSRIs are only effective for one-third of patients [27], highlighting the urgent need for new treatment methods. More and more evidence shows that exercise training, as a non pharmacological method, can increase Neurotrophin in the brain, thereby improving memory [28-30]. Studies have shown that moderate intensity swimming training [28,30]. Independent wheel exercise [28] and Treadmill training [29] can both inhibit the expression of neuroinflammation in adult [30] and elderly animals [29], which is consistent with the results of this study. After exercise training, the expression of inflammatory factors IL-1 and IL-6 in the gut and brain tissue decreased.

This study successfully established a PSD model using adult combined middle cerebral artery occlusion/reperfusion (MCAO/r) model and CUMS stimulation. The increase in sucrose intake in SPT and immobility in FST can both serve as indicators of depression. Consistent with previous reports [31], all behavioral and molecular biology tests have shown the relief effect of exercise training on depressive like behavior.

More and more research evidence suggests that the gut microbiota plays a crucial role in regulating brain function and human behavior. Inflammatory factors are important for the interaction between depression and Gut microbiota, and play an important role in regulating the brain gut axis. As is well known, gut microbiota, as initiators of micro inflammatory states, play an important role in regulating human immune processes. They are crucial for the establishment, development, and maintenance of the host immune system [32-33]. The changes in gut microbiota of depression model mice prepared with antibiotic dry preparation lead to changes in peripheral inflammation and the production of central inflammatory factors, thereby inducing depression [34]. For example, when the intestinal microbiota is destroyed, peripheral inflammatory factors can directly cross the Blood-brain barrier, or activate the HPA axis through the migration of inflammatory factors to increase the level of the stress hormone glucocorticoid, thereby reducing Brain-derived neurotrophic factor and inducing depression. The presence of central inflammatory factors was also observed in the brains of mice with depression models. This to some extent explains the midgut of this study

4. Conclusion

Exercise training can effectively improve the symptoms of post-stroke depression in rats and reduce the expression of inflammatory factors. However, the research on the relationship between the mechanism of Gut microbiota and PSD is relatively scarce. The existing research can only confirm that there is an internal relationship between post-stroke depression and the status of Gut microbiota, and the specific mechanism and treatment target still need to be further explored.

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Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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