Original Research Article

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Effect of aerobic intermittent exercise on the decreased cognitive ability induced by $PM_{2.5}$ exposure in rats

https://doi.org/10.1515/teb-2024-0009
Received May 7, 2024; accepted July 13, 2024;
published online August 2, 2024

Abstract

Objectives: This study aimed to investigate the protective effects and underlying mechanisms of aerobic intermittent exercise on cognitive impairment by $PM_{2.5}$ exposure.
Methods: Thirty-two rats were randomly divided into four groups: sedentary, exercise, sedentary + $PM_{2.5}$ exposure, and exercise + $PM_{2.5}$ exposure. The exercise groups underwent 8 weeks of exercise training (5 days of exercise per week). Subsequently, $PM_{2.5}$ exposure groups were subjected to $PM_{2.5}$ for three weeks. Post-exposure, we assessed cognitive abilities (shuttle box test), hippocampal tissue structure, related inflammatory factors (TNF-α, IL-6, IL-1β), the protein of inflammatory responses mechanism (P65, IκκB) and cognitive-related protein levels (BDNF, Aβ-42).
Results: $PM_{2.5}$ exposure caused cognitive impairment, abnormal histopathological changes, reduced cognitive related protein and increased pro-inflammatory cytokine levels. Analysis of shuttle box test data revealed significant main effects on the passive avoidance latency times measured in rats ($p<0.05$). Aerobic intermittent exercise improves spatial learning decline in rats induced by $PM_{2.5}$. Conversely, the Exercise + $PM_{2.5}$ group demonstrated a significant reduction in latency of 24.9 % compared to the Sedentary + $PM_{2.5}$ group ($p<0.05$, ES=1.41).
Conclusion: Aerobic intermittent exercise may help in protecting against the decrease of cognitive ability induced by $PM_{2.5}$ exposure.

Keywords: aerobic interval exercise; particle matters; cognitive behavior abilities; NFκB signal pathway

Introduction

Particulate matter with diameters less than 2.5 μm ($PM_{2.5}$) can be inhaled through the respiratory system due to their small aerodynamic size, which allows them to enter the body and pose serious health risks [1].

The deposition of particulate matter into the lungs can induce localized inflammation, leading to respiratory disorders such as asthma and chronic lung obstruction [2]. It is worth noting that it has been reported that fine particulate matter can target the respiratory tract to initiate sequential effects, activate local inflammatory factors to trigger an inflammatory cascade, leading to neurological damage and seriously threatening cognitive function [3, 4]. Studies have shown that air pollutants can trigger oxidative stress and inflammation, reducing the expression of brain-derived trophic factors and disrupting neurotransmitter transmission, ultimately leading to cognitive decline [5, 6].

Neurodevelopment of fetuses exposed to $PM_{2.5}$ during pregnancy is affected, and adolescents living in $PM_{2.5}$-polluted environments for long periods of time have significantly reduced learning and memory abilities [7]. Middle-aged and elderly people who live in polluted environments for long periods of time are significantly more likely to suffer from neurodegenerative diseases [8]. Reduce damage of $PM_{2.5}$ to the cognitive function of the organism should receive wide attention, provided that the air pollution situation has not been completely controlled.

Exercise has become a widely accepted economical, non-invasive and effective intervention for the prevention

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of chronic metabolic illnesses [9]. Expression of oxidative stress factor in the hippocampus of high-fat dietary obese mice was significantly decreased after six weeks of aerobic intermittent exercise intervention; in the Morris water maze test, the avoidance latency of the rats in high-fat exercise group shorter than that in the sedentary and high-fat group, indicating that the spatial memory ability of the mice was improved and the cognitive decline was improved after six weeks of aerobic interval exercise [10–12]. Aerobic interval exercise for two months in 20-month-old rats significantly increased BDNF levels in the hippocampus and prefrontal lobes, and the rats performed better than the control rats in behavioral tests, and aerobic interval exercise can effectively improve the spatial learning ability of aged rats [13]. The project team's previous study also found that regular 8-week aerobic interval exercise can alleviate the damage caused by $PM_{2.5}$ to the body's lungs to a certain extent [14]. However, there remains a lack of basic experimental evidence supporting the protective effects of exercise on $PM_{2.5}$-induced cognitive impairment and its mechanisms, necessitating further exploration [1]. Therefore, this study aims to evaluate the protective mechanisms of aerobic intermittent exercise against $PM_{2.5}$-induced cognitive damage [14]. The hypothesis is that intermittent exercise will mitigate cognitive damage caused by $PM_{2.5}$ exposure. The summary of this article is presented in Figure 1.

Materials and methods

Animals

Thirty-two 8-week-old male Wistar rats were purchased from Beijing Vital River Laboratory Animal Technology Co. Ltd. [14]. The rats were housed in separate cages with five rats per cage, in strict accordance with the guidelines for rodent experimental animal care. They had free access to food and water, with bedding changed regularly. Each cage provided 500 mL of water through a drinking bottle and measured the amount of water consumed daily. In addition, rat feed pellets were weighed and consumption recorded. The length and weight of the rats were measured periodically during the feeding period. The physical condition of the rats was observed during this period to see if they were injured or sick [15]. The animal room was maintained at a temperature of 23 ± 1 °C, humidity at 45–55 %, and a 12-h light/dark cycle [14]. All procedures were approved by the Animal Ethical Committee of China Institute of Sports Science (approval number: CISSLA-2017003) [14].

Experimental design

The rats were randomly divided into four groups: sedentary (S), exercise (E), sedentary + $PM_{2.5}$ exposure (S + PM), and exercise + $PM_{2.5}$ exposure (E + PM) [14]. The exercise groups underwent an eight-week aerobic interval treadmill exercise, followed by $PM_{2.5}$ exposure for the PM-related groups [14]. The exposure duration was 6 h per day [14]. Immediately after exposure, animal behavioral tests were conducted. Finally, the rats were anesthetized via intraperitoneal injection, and hippocampal tissues were collected [14].

Exercise program

In this study, rats were tested using a rat four-channel exercise treadmill (Columbus, USA), and the rat four-channel metabolic monitoring system (Columbus, USA) was used to collect and analyze the changes in oxygen uptake and maximum oxygen uptake during the test, and to determine the corresponding intensities of the rats' exercise intensity in the exercise experiments [16]. After 3 days of acclimatization feeding, rats in the E and E + $PM_{2.5}$ groups underwent acclimatization treadmill training [17]. After acclimatization, 8 weeks of exercise training (5 days of exercise per week) were performed. The exercise program was as follows: warm-up for 5 min at a speed of 12 m/min (50–55 % VO_2max); accelerate to 40 m/min (80–90 % VO_2max) for 4 min after completing the warm-up; then decelerate to 15 m/min (65–70 % VO_2max) for 3 min. The formal exercise phase alternated between 40 m/min and 15 m/min cycles for a total of seven cycles, ending with a 5 min finishing recovery exercise at 12 m/min [14, 17, 18]. We assessed VO_{2max} to adjust the practical running speed and set the exercise intensity, which was evaluated with the Columbus Oxymax Lab Animal Monitoring System and an animal treadmill chamber (Columbus, USA) every 2 weeks.

$PM_{2.5}$ exposure system

The $PM_{2.5}$ Animal Whole Body Inhalation Exposure System (Beijing Huironghe Technology Co., Ltd.) was used to collect and concentrate outdoor air, which was then discharged into the exposure chamber [14]. The system located at Tongzhou District, Beijing, China. The particulate exposure system in this study collected outdoor particulate matter in real time, and by concentrating and enriching the collected particulate

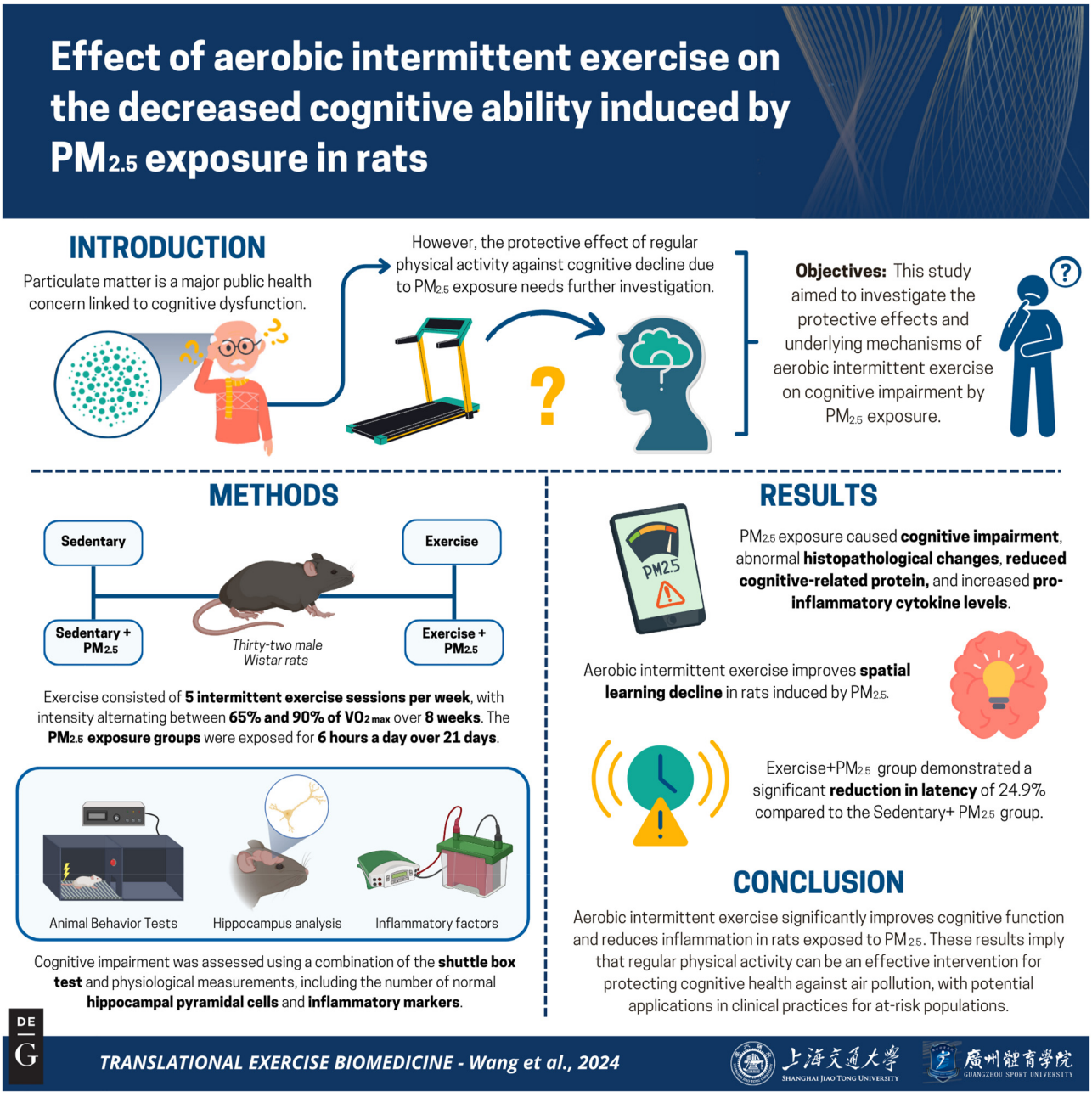

Figure 1: Graphical representation of this article. Figure created with BioRender.

matter in the exposure chamber, whole-body exposure inhalation was performed on rats, which is close to the real-life outdoor inhalation situation of human beings. Air quality data, including PM_{10}, NO_2, CO, SO_2, and O_3 concentrations, were recorded from the Ministry of Ecology and Environment of China website [14]. Exposure phase started in mid-October 2018 and lasted for 21 days, with each session lasting 6 h. Groups S and E were kept under normal conditions during this period.

Components in chamber collected by 47 mm Teflon filter, analyzing PAHs and metals. Sample extracted by accelerated reaction system and purified using silica/alumina columns [14, 19, 20]. PAHs were analyzed by gas chromatography coupled with mass spectrometry (Agilent 5890A and 5975C). The 16 quantified PAHs included naphthalene (NAP), acenaphthene (ACE), fluorene (FLO), acenaphthylene (ACY), phenanthrene (PHE), fluoranthene (FLA), anthracene (ANT), pyrene (PYR), benzo(k)fluoranthene (BkF) [14]. The

concentrations of Cr, Cu, Zn, Cd, Ni, Mn, Hg, Pb, and As were determined using inductively coupled plasma mass spectrometry (ICP-MS, DIONEX, United States) [14].

Animal behavior tests

Immediately after the PM-related groups completed the exposure, we performed behavioral experiment of the rats by shuttle box (Beijing Zongshi Technology Co., Ltd., China). During the formal experiment, the rats were placed in either area of the shuttle box and acclimated to the environment for 5 min, after which the rats were given acoustic and optical stimuli and electrical stimuli at the same time, and the time for the rats to escape from the shuttle box was recorded, which was used to evaluate and analyse the cognitive ability. The time the rats spent looking for escape after receiving acoustic and electric stimuli, the escape latency was tested. The escape latency of different rats was recorded and the cognitive ability of the rats was judged.

Pathological analysis of the hippocampus

Tissues were fixed in neutral formaldehyde fixative for 48 h. Samples were made into sections (4 μm) by gradient alcohol dehydration, xylene transparency, and paraffin embedding. Three consecutive slices were taken for mounting. The slices were baked in a constant temperature drying oven at 65 °C for 1.5 h. Using paraffin embedding technique and HE staining technique, Leica light microscope was used for observation and photographing, and the histological changes in the cornu ammonis 1 (CA1) area of the hippocampal region of the rats were observed at 40, 100, and 400 *X* of the field of view, respectively. The number of normal hippocampal pyramidal cells (meaning pyramidal cells with large, round nuclei, light and clear hematoxylin staining, distinct nucleoli and membranes, and abundant cytoplasm) in each section was counted.

Western blotting

Proteins extracted from hippocampal, tested for inflammatory factors (TNF-α, IL-6, IL-1β), NFκB pathway-related proteins (IκκB, P65), as well as Aβ-42 and BDNF. Concentrations were determined using the BCA. According to the molecular weight of the target protein, 12 % separating gel was prepared and the concentration of concentrated gel was 5 %. Protein samples to be detected were loaded with 20 μg/well. Primary antibodies used included IκκB (CST, 1:1,000), TNF-α (Abcam, 1:1,000), P65 (CST, 1:2,000), IL-6 (Immunoway, 1:1,000), BDNF (Abcam, 1:1,000), Aβ-42 (SANTA, 1:500), and IL-1β (Abcam, 1:4,000) [21, 22]. Detection was performed with an ECL kit [14].

Statistical analysis

Data were expressed as mean ± standard deviation (Mean ± SD) and analyzed using SPSS software (version 20.0, IBM SPSS Statistics, Chicago, IL, USA). Normal distribution and chi-square tests were performed, followed by two-way ANOVA to analyze main and interaction effects. If significant interactions were found, one-way ANOVA was used to examine simple main effects, with LSD post-hoc tests for pairwise comparisons [23, 24]. Significance was set at $p<0.05$. Cohen's d effect size (ES) was calculated, with ES≥0.80 as high, 0.80>ES≥0.50 as medium, and 0.50>ES≥0.20 as small [25].

Results

Concentrations and composition of inhalable particulate matter exposure

After an 8-week exercise training regimen, rats in the S + $PM_{2.5}$ and E + $PM_{2.5}$ groups underwent whole-body exposure to $PM_{2.5}$ in a controlled environment. The whole-body inhalation exposure system for $PM_{2.5}$ was produced by Beijing Huironghe Science and Technology Co. Ltd.. This system employs an air concentration enrichment method that actively collects outdoor air, concentrates it, and directs it into the exposure chambers. The exposure phase commenced in mid-October 2018 and continued for 21 consecutive days. Each exposure session lasted 6 h, during which the rats were maintained under normal husbandry conditions. Exposure concentrations were recorded throughout, with a mean daily concentration of 237.01 ± 206.41 μg/m^3 (ranging from a high of 651.70 ± 70 μg/m^3 to a low of 21 ± 41 μg/m^3). The S and E groups of rats were also maintained normally during this period. There was no difference in the types of outdoor air pollutants between the ambient environment and inside the exposure chambers. The three most prevalent metal elements detected were zinc, manganese, and copper. The concentrations of compounds and metals within the exposure chambers showed in Table 1 [14].

Protective effect of exercise on $PM_{2.5}$-induced cognitive decline

Shuttle box test data revealed significant main effects of rats on the passive avoidance latency times measured in rats

Table 1: Mean concentrations compounds and metallic substances in exposure chamber [14].

Metals	Mass concentration ($ng \cdot m^{-3}$ $PM_{2.5}$)	PAHs	Mass concentration ($\mu g \cdot m^{-3}$ $PM_{2.5}$)
Cr	11.41	Naphthalene	0.127
Mn	68.15	Acenaphthylene	0.033
Ni	4.80	Acenaphthene	1.108
Cu	21.91	Fluorene	1.145
Zn	216.38	Phenanthrene	2.195
As	12.32	Anthracene	0.145
Cd	2.94	Pyrene	0.293
		Fluoranthene	0.183
		Chrysene	0.052
		Benzo(a)anthracene	0.115
		Benzo(b)fluoranthene	0.207
		Benzo(k)fluoranthene	0.067
		Benzo(a)pyrene	0.102
		Benzo (g,h,i)perylene	0.148
		Indeno (1,2,3,c,d)Pyrene	0.153
		Dibenz (a,h) Anthracene	0.052

($p<0.05$). This indicates that both exercise and $PM_{2.5}$ exposure significantly influence passive avoidance latency. Between-group comparisons showed that the S + $PM_{2.5}$ group compared to the S group, the passive avoidance latency increased by 21.0 % ($p>0.05$, *ES*=0.86). Relative to the E group, the S + $PM_{2.5}$ group exhibited a significant increase in latency of 50.1 % ($p<0.05$, *ES*=2.11). Conversely, the E + $PM_{2.5}$ group demonstrated a significant reduction in latency of 24.9 % compared to the S + $PM_{2.5}$ group ($p<0.05$, *ES*=1.41).

Hippocampal CA1 region of the S, E group rats, cells were densely packed with clear, well-defined layers, and cell morphology was intact with large, round nuclei. In the S + $PM_{2.5}$ group, however, cells in the hippocampal CA1 area were sparsely arranged with notable vacuolization and gaps between cells, incomplete cellular morphology, blurred edges, and shrunken nuclei. The E + $PM_{2.5}$ group showed denser cell arrangements and more intact structural morphology in the hippocampal CA1 area compared to the S + $PM_{2.5}$ group, with an increase in cell count and clear cell borders (Figure 2).

Compared to the S group, protein levels of hippocampal BDNF the S + $PM_{2.5}$ group decreased by 19.1 % ($p>0.05$, *ES*=0.84). Significant decrease in BDNF protein levels by 20.9 % in the S + $PM_{2.5}$ group compared to the E group ($p<0.05$, *ES*=1.34). In contrast, the E + $PM_{2.5}$ group increased significantly by 26.5 % compared to the S + $PM_{2.5}$ group ($p<0.05$, *ES*=0.95).

Aβ42 protein levels in hippocampal tissue, there was an increase of 10.8 % in the S + $PM_{2.5}$ group compared to the S group ($p>0.05$, *ES*=0.61). The Aβ42 level significantly rose by 18.1 % in the S + $PM_{2.5}$ group compared to the E group ($p<0.05$, *ES*=1.05). However, the E + $PM_{2.5}$ group showed a decrease in Aβ42 levels by 12.5 % compared to the S + $PM_{2.5}$ group ($p>0.05$, *ES*=0.94) (Figure 3).

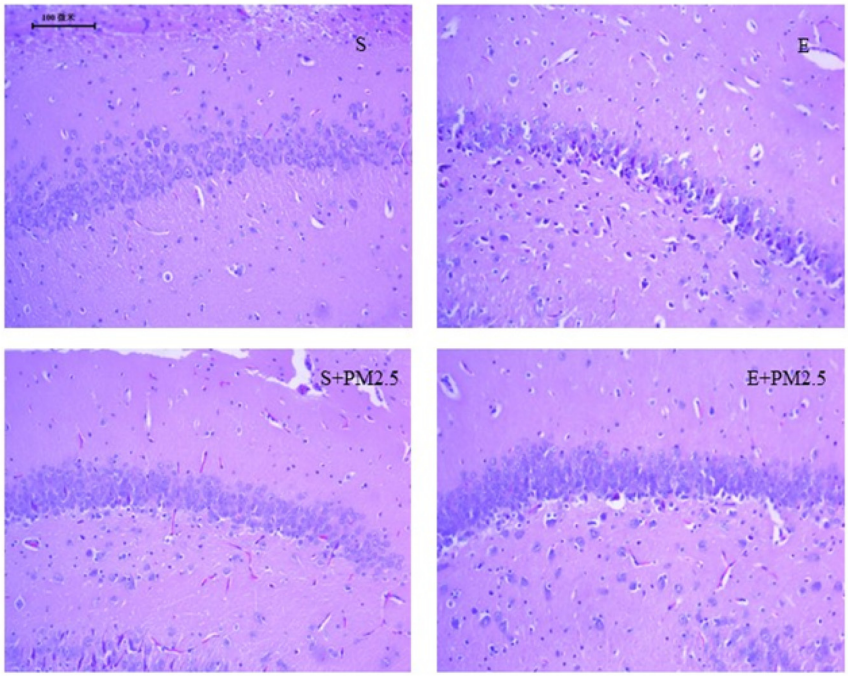

Figure 2: Photomicrographs of rat hippocampal CA1 region (HE), n=6.

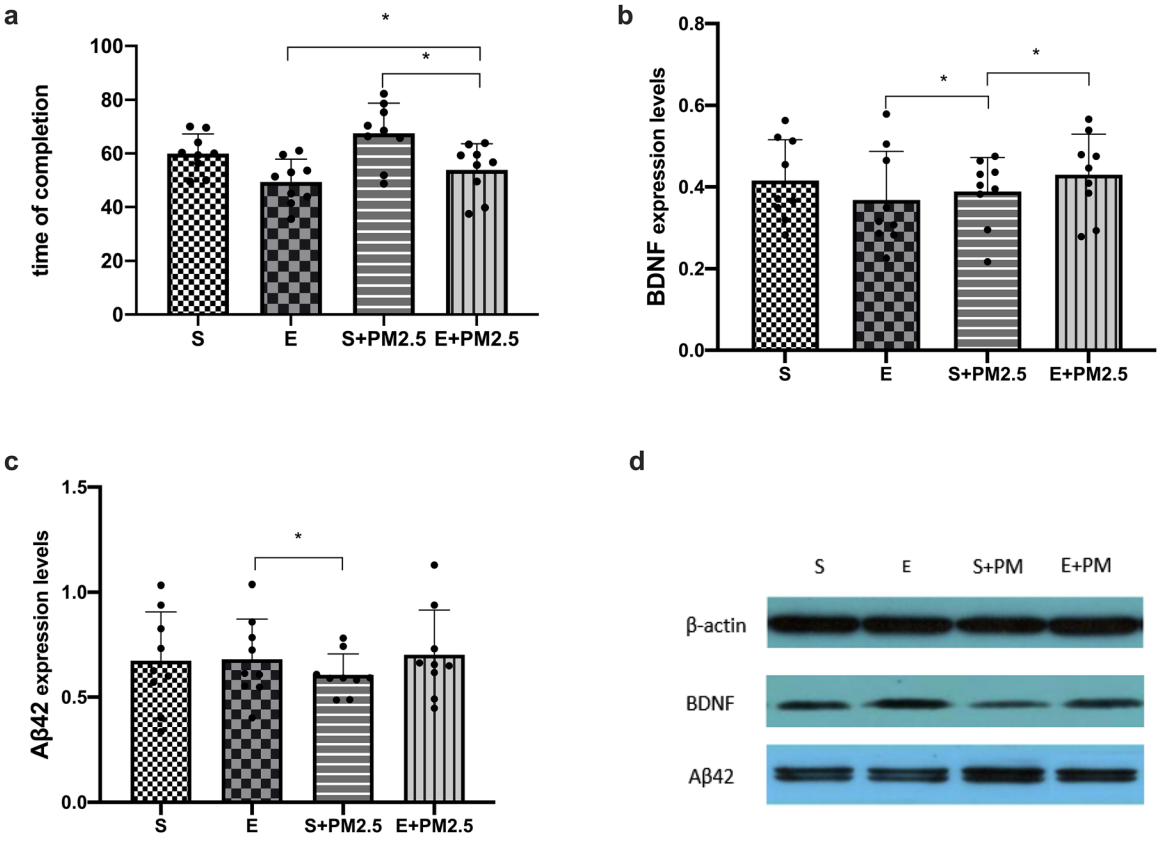

Figure 3: Protective effects of exercise on cognitive decline caused by $PM_{2.5}$. (a) Passive avoidance latency times. (b and c) Protein levels of BDNF and Aβ42 in hippocampal tissue. (d) Protein binging pattern determined by western blot. Data represent mean ± SD, n=9, *p<0.05 between groups.

Mechanisms of exercise's protective effects against cognitive decline induced by inhalable particulate matter

In the hippocampal tissue, TNF-α protein expression in the S + $PM_{2.5}$ group increased by 21.2 % compared to the S group (p>0.05, ES=0.87) and significantly by 29.0 % compared to the E group (p<0.05, ES=1.12). Conversely, the E + $PM_{2.5}$ group showed a 29.0 % decrease compared to the S + $PM_{2.5}$ group (p<0.05, ES=1.12).

IL-1β protein expression in the S + $PM_{2.5}$ group was 22.0 % higher than in the S group (p<0.05, ES=0.86) and significantly higher by 22.0 % compared to the E group (p<0.05, ES=0.97). In the E + $PM_{2.5}$ group, IL-1β expression increased by 16.0 % compared to the S + $PM_{2.5}$ group but was not statistically significant (p>0.05, ES=0.74).

IL-6 protein expression in the E group was reduced by 26.1 % compared to the S group (p<0.05, ES=1.50). In the S + $PM_{2.5}$ group, IL-6 levels significantly higher than in the S group by 20.7 % (p<0.05, ES=1.90) and higher by 52.2 % compared to the E group (p<0.05, ES=3.79). In the E + $PM_{2.5}$ group, IL-6 levels were significantly reduced by 20.7 % compared to the S + $PM_{2.5}$ group (p<0.05, ES=0.88) and by 31.4 % compared to the S + $PM_{2.5}$ group (p<0.05, ES=2.14).

P65 protein expression in the S + $PM_{2.5}$ group increased by 11.1 % compared to the S group (p>0.05, ES=0.49) and significantly by 22.0 % compared to the E group (p<0.05, ES=1.63). In the E + $PM_{2.5}$ group, P65 protein levels decreased by 11.1 %, but this was not statistically significant (p>0.05, ES=1.05).

IκκB protein expression in the S + $PM_{2.5}$ group was 30.4 % higher than in the S group (p<0.05, ES=1.07) and 36.4 % higher than in the E group (p<0.05, ES=1.33). In the E + $PM_{2.5}$ group, IκκB protein levels decreased by 23.3 % compared to the S + $PM_{2.5}$ group (p<0.05, ES=0.99) (Figure 4).

Discussion

Air pollution-induced cognitive decline has been widely recognized, while the ability of regular aerobic exercise to alleviate cognitive decline has been confirmed in clinical study [26]. Long-term regular exercise could mitigate cognitive decline induced by air pollution. It was found that

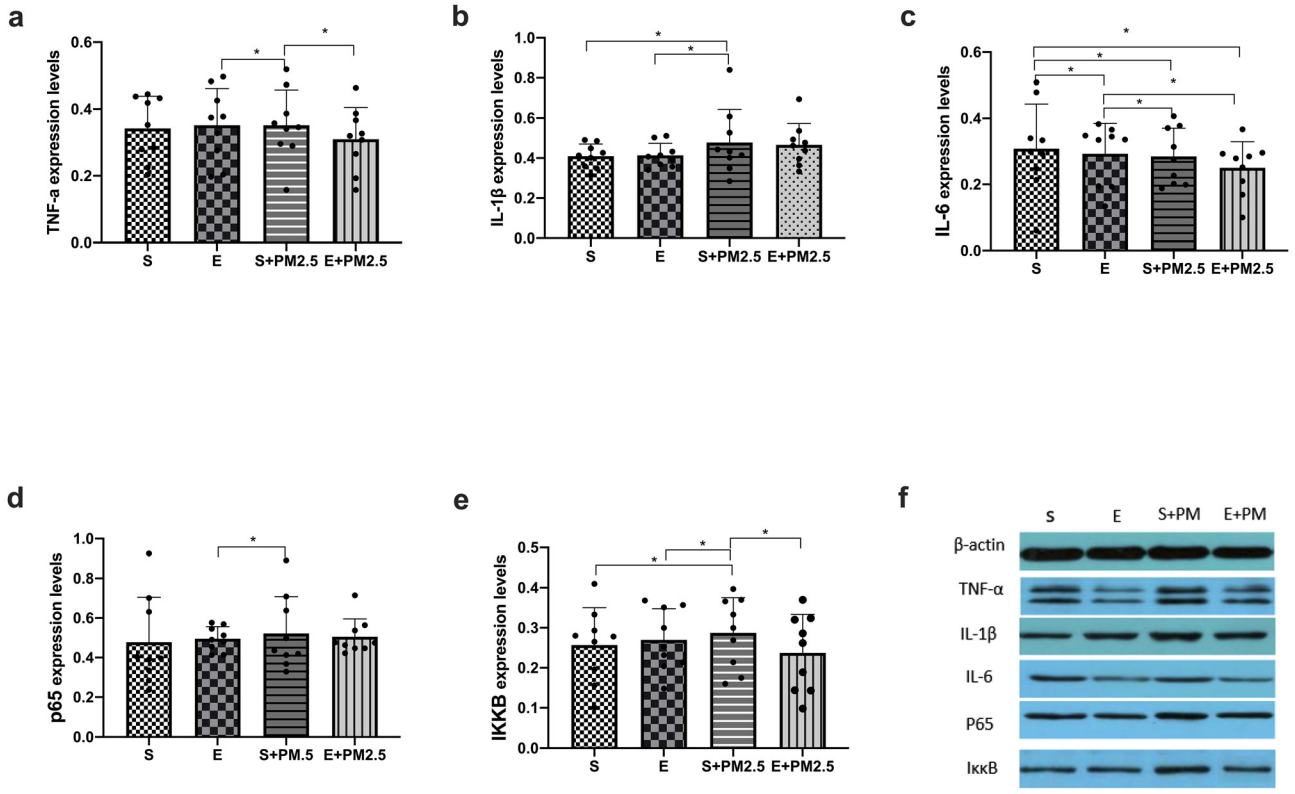

Figure 4: Mechanisms of exercise's protective effects against cognitive decline induced by $PM_{2.5}$. (a–c) protein levels of pro-inflammatory cytokines (TNF-α, IL-1β and IL-6) in hippocampal tissue of rats. (d–e) proteins levels of P65 and IκκB in hippocampal tissue. (f) Protein binging pattern determined by western blot analysis. Data represent mean ± SD, n=9, *p<0.05 between groups.

aerobic interval training effectively prevents $PM_{2.5}$-induced cognitive decline in rats and may exert its protective effects by reducing inflammation through the NFκB signaling pathway [14, 17]. These findings provide theoretical and experimental support for using exercise to counteract $PM_{2.5}$-induced cognitive impairment.

A pre-exercise model was constructed by aerobic intermittent exercise over a period of 8 weeks, and the human body was simulated in an air-polluted environment by airborne particulate matter exposure. The whole cycle of exposure was 22 days, each exposure lasted 6 h and the average concentration was 237.01 ± 206.41 μg/m^3. According to the $PM_{2.5}$ concentration level index for division, the exposure cycle reached severe pollution (250–500 μg/m^3) for 8 days, heavy pollution (150–250 μg/m^3) for 4 days, moderate pollution (50–150 μg/m^3) for 5 days, light pollution (35–50 μg/m^3) for 2 days, and excellent air quality for 3 days, this air pollution. The pattern of change is similar to the previous trend of the more polluted cities in the north of China (Beijing-Tianjin wing area) in early winter [27]. The real-time collection of outdoor air in Beijing, the air particulate matter concentration fluctuates in a wide range due to environmental influences, but it is also in the normal fluctuation.

In this study, we analyzed the compositional content of $PM_{2.5}$ in the exposure chamber and found that the PAHs in the exposure bin were dominated by phenanthrene, along with benzene and pyrene. The study showed that exposure to phenanthrene causes abnormalities in nerve fiber and induces nerve fiber disorders [28]. Study has shown that the PAH compounds benzene and pyrene can cause neuronal damage through oxidative stress and induce degenerative neuronal lesions [29]. In this study, Zn, Mn and Cu were the three most abundant heavy metals in the exposure chamber by ICP-MS. Study has shown that intake of Mn, Cu can be toxic to the nervous system and contribute to neurodegeneration [30, 31]. Elevated levels of Cu are associated with mild cognitive impairment and Alzheimer's disease processes, and Parkinson's disease is associated with elevated Mn [32, 33]. Study has shown that increases in Mn and Cd lead to cognitive decline in older adults [34]. It has been found that the presence of polycyclic aromatic hydrocarbons (PAHs) and heavy metals in air pollution particulate matter produced by vehicle exhausts [35]. Study has shown that in cognitive assessments of children, each increase of 5 μg/m^3 in particulate matter concentration reduces performance on cognitive tests [36]. Study has shown that each

1 μg/m^3 increase in $PM_{2.5}$ during outdoor exercise diminishes the cognitive benefits of interval exercise, as assessed by cognitive function [37]. The exposure chamber in this study was located near several highways and urban arterial roads, and it was hypothesized that the main source of $PM_{2.5}$ in this environment was motor vehicle exhaust, suggesting that traffic-sourced air pollutants may be closely related to brain health, and that the duration of outdoor activities during peak traffic hours should be minimized.

Neurons maintain projection and transmission links within the hippocampus and between other brain regions, and thus the hippocampus is considered to be an important region in the formation of learning and memory processes [13, 38]. The hippocampus is an important region that play a role in learning and memory processes, as well as in the transmission of spatial localization in the animal brain, and it has been found that there are positional cells within the hippocampus, as well as the ability to identify environmental and spatial information [39, 40]. In the present study, it was found that after particulate matter exposure, the S + $PM_{2.5}$ exposed group showed sparse cellular arrangement, incomplete cellular morphology, fuzzy edges, broken and crumpled nuclei in the hippocampal region of the rats as compared to the control group. Previous studies have also found impaired spatial learning memory ability in mice by co-exposure to various types of air pollutants such as $PM_{2.5}$, sulphur dioxide and nitrogen dioxide, and observation of the mouse brain by morphological tests revealed the presence of inflammatory cell infiltration and neuronal cell damage existed in the mice. Oxidative stress induced by long-term exposure to air pollution has also been found to cause a reduction in the number of neurons in the CA1 region of the rat brain, and may be associated with a decline in performance on behavioral tests [41, 42]. After 8 weeks of aerobic intermittent exercise, the hippocampal tissue structure of the E + $PM_{2.5}$ rats was more neatly arranged, regular, and clearly defined than that of the S + $PM_{2.5}$ rats, and the cellular structure was more complete. It was found that continuous aerobic exercise could effectively improve the lesions of the damaged hippocampal tissue and reduce the number of apoptotic cells, thus improving the learning and memory ability of mice [43, 44].

Brain Derived Neurotrophic Factor (BDNF) is an important signaling factor that promotes and maintains synaptic plasticity in the hippocampus and Long-term Potential (LTP), which plays an important role in the maintenance of cognitive performance [13, 43, 45]. Aβ42 is a widely studied pathological marker currently used in clinical practice for the assessment of attention deficit disorder and is the primary method of assessing Alzheimer's disease [46, 47]. Elevated levels of Aβ42 promote the progression of Alzheimer's disease, causing degenerative cognitive pathology. Research show that people with Alzheimer's disease have abnormalities in brain activity that are caused by the effects of Aβ42 on neurons and lead to impaired cognitive function [48]. Study has shown that upregulation of Aβ42 levels, in addition to causing neurological damage, can adversely affect the cerebral vasculature, further brain health [49]. Hippocampal tissue in the brain is important for memory function and spatial navigation due to its role in encoding and recall of experiences [50]. The CA1 region in the hippocampus is located on the side of the main pathway connecting the hippocampus to the cortex, which is rich in synapses and plays an important role in memory formation [51]. CA1 pyramidal neurons also function in the formation of self-awareness and the integration of multimodal sensory information about environmental stimuli during locomotion [52, 53]. Hippocampal volume is correlated with response to physical activity as influenced by hippocampal cell plasticity [54]. Study shows that being physically active can boost hippocampal neural production, leading to improved cognitive functioning [53]. Hippocampal volume was found to be significantly larger in older adults after completing a 1-year aerobic exercise intervention in a population of cognitively normal older adults [53, 55].

After 8 weeks of aerobic intermittent exercise, BDNF levels in the E + $PM_{2.5}$ group significantly increased compared to the S + $PM_{2.5}$ group ($p<0.05$, ES=0.95) [21]. Additionally, hippocampal tissue Aβ42 levels were down-regulated in the E + $PM_{2.5}$ group compared to the S + $PM_{2.5}$ group ($p>0.05$, ES=0.94) [24]. Previous studies on stroke improvement through exercise showed that after 4 weeks of treadmill training, hippocampal BDNF levels were significantly higher in the exercise group of Wistar rats compared to the control group [56].

After 8 weeks of training, 4 days a week, rats injected with Aβ42 had improved cognitive memory and decreased abnormal β-amyloid deposits in brain tissue, following aerobic and resistance training [57]. Hippocampal tissue Aβ42 levels were significantly reduced in C57BL/6 mice following an 8-week running platform aerobic training intervention [58]. Combined with the escape time of rats in the shuttle box test it can be found that an 8-week aerobic intermittent pre-exercise reduces the escape time and improves the spatial learning cognitive ability of rats, which is consistent with previous studies. This may be related to the alleviation of BDNF down-regulation and inhibition of Aβ42 elevation after regular pre-exercise, which in turn improves neuronal degenerative lesions in the hippocampal region and ameliorates inflammatory infiltration. Therefore, the 8-week aerobic intermittent pre-exercise used in this study can effectively improve spatial learning memory loss and cognitive decline caused by $PM_{2.5}$ inhalation.

Relevant studies have shown that $PM_{2.5}$ particulate matter can induce systemic inflammation by translocating and causing inflammatory responses in various tissues [59]. Inflammatory reactions in the brain and central nervous system can severely impair cognitive functions. Notably, IL-6 and TNF-α levels were significantly elevated in the brain tissues of Alzheimer's disease model rats with severe dementia, leading to mitochondrial swelling [41, 60].

Through 8 weeks of aerobic intermittent pre-exercise, hippocampal TNF-α and IL-1β protein levels were downregulated, and IL-6 protein levels were significantly decreased in the E + $PM_{2.5}$ group compared to the S + $PM_{2.5}$ group [61]. Previous studies have demonstrated that aerobic exercise can effectively reduce elevated levels of inflammatory factors like TNF-α and IL-1β in the hippocampus, prefrontal lobe, and other brain tissues due to $PM_{2.5}$ inhalation [6, 62, 63]. In this study, 8 weeks of aerobic interval exercise effectively reduced TNF-α, IL-1β, and IL-6 protein levels in the hippocampal tissues of rats exposed to $PM_{2.5}$, mitigating neuropathy caused by these inflammatory factors and potentially improving cognitive abilities.

The NFκB signaling pathway, crucial for cell proliferation, apoptosis, immune response, and oxidative stress, plays a significant role in this process [64–66]. IκκB, an important transcription factor in the NFκB pathway, is closely related to its classical pathway [67, 68]. In this study, 8 weeks of aerobic intermittent pre-exercise significantly reduced IκκB protein levels and P65 protein levels in the hippocampal tissues of the E + $PM_{2.5}$ group compared to the S + $PM_{2.5}$ group. Previous studies have shown that an 8-week running training intervention in Alzheimer's disease model rats resulted in decreased hippocampal levels of TNF-α and IL-1β, as well as reduced phosphorylation levels of P65 and IκκB [9, 61, 69, 70].

These findings suggest that aerobic exercise can effectively inhibit P65 and IκκB expression, thereby blocking the activation of the NFκB pathway, reducing the inflammatory response, and preventing neuropathy. Consequently, this can improve cognitive function. In summary, this study found that 8 weeks of aerobic intermittent pre-exercise can inhibit P65 and IκκB expression, antagonizing the activation of the NFκB pathway, reducing neural inflammation, and potentially improving cognitive decline [1].

In this study, after an 8-week aerobic intermittent pre-exercise intervention, it was found that regular exercise fitness training before the start of the haze cycle environment could effectively resist the downward trend of cognitive function caused by haze to a certain extent and inhibit the development of inflammation in the hippocampal region. Based on the above analysis, the research hypothesis is proposed that long-term regular aerobic exercise pre may improve cognitive function by resisting atmospheric particulate matter-induced inflammatory factors in hippocampal tissues, and then inhibiting the NFκB inflammatory signaling pathway. In addition, the above results also suggest that although pre-exercise can resist the cognitive impairment caused by particulate matter to a certain extent, the relevant study on the safe exposure value of particulate matter still need to be further verified in the future, thus suggesting that we should reasonably adjust our travelling plan according to the concentration of air pollutants, control the time of outdoor activities and work, and take proper physical protection measures.

There are also some limitations of this study. The present study shows that mitigation of aerobic interval exercise for cognitive decline induced by subphrenic airborne particulate matter exposure. However, there is a lack of reference for groups subjected to chronic air exposure, especially those who have been exposed to air pollution for a long period of time during pregnancy and early childhood development [71]. Meanwhile, related study have shown that cognitive decline induced by air pollution is more prevalent in the elderly population, whereas this paper uses 8-week-old rats and does not address the response to changes in cognitive function at different ages [72]. Lastly, due to the seasonal and cyclical nature of smog weather, this study first opted for 8-week regular physical exercise, followed by subacute $PM_{2.5}$ exposure. This approach aims to verify the protective effect of exercise habits against cognitive decline caused by air pollution. Future experiments will explore other interaction effects between exercise and air pollution (e.g., Chronic $PM_{2.5}$ exposure followed by exercise intervention).

Conclusion

Aerobic intermittent exercise significantly alleviated cognitive impairment induced by PM2.5 exposure in rats. This exercise likely exerts positive effects on cognitive function by reducing inflammation and protecting hippocampal structure. This study provides scientific evidence for understanding the negative impact of air pollution on cognitive function and supports aerobic intermittent exercise as an effective non-pharmacological intervention to mitigate the health risks associated with air pollution.

Research ethics: The animal experiments involved in this study were approved by the Animal Experiment Ethics Committee of the China Institute of Sport Science. The approval number is CISSLA-2017003. This study adhered to the Declaration of Helsinki as well as national standards concerning the welfare and ethics of experimental animals.

Informed consent: Not applicable.
Author contributions: Conceptualization: Zhongwei Wang, Fei Qin, Jiexiu Zhao. Data curation: Zhongwei Wang, Fei Qin, Dongzhe Wu. Formal analysis: Zhongwei Wang, Dongzhe Wu. Funding acquisition: Jiexiu Zhao. Methodology: Zhongwei Wang, Fei Qin. Project administration: Jiexiu Zhao. Visualization: Zhongwei Wang, Dongzhe Wu. Writing – original draft: Zhongwei Wang, Fei Qin. Writing – review & editing: Zhongwei Wang, Fei Qin.
Competing interests: The authors state no conflict of interest.
Research funding: This study was supported by project 24-28 supported by the Fundamental Research Funds for the China Institute of Sport Science, the National Natural Science Foundation of China (31900845 and 11775059).
Data availability: Data will be available upon request to the first author.

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