*Int. J. Morphol.,*
*42(3)*:614-622, 2024.

# Investigating Morphological Changes of the Hippocampus After Prolonged Aerobic Exercise in Mice: Neural Function and Learning Capabilities

## Investigación de los Cambios Morfológicos del Hipocampo Después De un Ejercicio Aeróbico Prolongado en Ratones: Función Neuronal y Capacidades de Aprendizaje

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**HUA, Z. & SUN, J.** Investigating morphological changes of the hippocampus after prolonged aerobic exercise in mice : Neural function and learning capabilities. *Int. J. Morphol., 42(3)*:614-622, 2024.

**SUMMARY:** Both the academic and popular worlds have paid close attention to the link between exercise and cognitive performance. It is increasingly important to understand the numerous mechanisms by which exercise might influence cognitive abilities in view of the continuous societal issues caused by aging populations and the prevalence of disorders associated to cognitive decline. A rising amount of evidence showing a favorable association between physical activity and cognitive well-being serves as the foundation for the justification for studying the effects of exercise on cognitive function and learning ability. The study employed an 8-week treadmill based on exercise on male adults C57BL/6 mice. The exercise group were engaged in 5 sessions a week gradually increasing the intensity of the protocol by 5 % each week. The Mice cognitive assessments were done using Morris Water Maze and Novel Object Recognition tests. The long term-impact on learning ability were further assessed through immmohistochemistry and molecular analysis of the hippocampal and prefrontal cortex tissues of the animals' brain tissues. The findings showed improved spatial learning abilities, recognition memory, and heighted synaptic plasticity indicated by elevated synaptic makers. The study underscores the role of long-term aerobic exercise in augmenting cognitive performance. It not only contributes to the understanding of the interplay between neuroplasticity and cognitive benefits but also the growing body of research on the impact of exercise on cognitive function.

**KEY WORDS: Long-Term Aerobic Exercise; Cognitive Function; Learning Ability; Neuroplasticity; Spatial Learning.**

## INTRODUCTION

Background and Rationale. Memory, attention, reasoning, and learning are just a few of the cognitive skills that might suffer from cognitive decline, which is caused by a variety of intricate circumstances. These deficits frequently play a role in the emergence of crippling diseases like dementia and Alzheimer's. Preventive measures are become more commonplace due to the lack of a permanent cure for many disorders. Exercise, particularly aerobic exercise, has been shown to have the potential to slow cognitive decline without the need of drugs. Animal studies have provided important insights into the physiological processes that connect exercise to cognitive performance. These include enhancing synaptic plasticity, stimulating neurogenesis, and increasing blood supply to the brain.

Additionally, using animal models allows researchers to control factors efficiently, which is essential for examining the causal relationship between exercise and cognitive outcomes. Numerous studies involving human participants have consistently shown a positive relationship between physical activity and cognitive health. However, these studies usually struggle to adequately control for potential confounding factors such as dietary habits, inherited traits, and socioeconomic status.

The Effects of Aerobic Exercise on Brain Health and Cognitive Function. The potential impact of aerobic exercise on cognitive function and brain health has been the subject of extensive investigation. Continuous, rhythmic motions that raise the heart rate and increase oxygen consumption are what define aerobicexercise. Multiple investigations have shown the effects of activities like jogging, swimming, and cycling on cognitive outcomes, illuminating the underlying mechanisms through which aerobic exercise confers its

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Received: 2024-01-31 Accepted: 2024-03-09

cognitive benefits. In a study conducted in 2006, Ikeda *et al*. (2006), showed a correlation between therapies including aerobic exercise and improved cognitive performance, particularly in tasks involving executive functioning, attention, and memory. Additionally, it has been discovered that aerobic exercise causes circulatory changes that improve vascular health. The aforementioned phenomena has important ramifications for brain health since it encourages higher cerebral blood flow, making it easier to transport oxygen and important nutrients to the brain consistently (Liu *et al*., 2011).

Moreover, aerobic exercise has been demonstrated to increase neurogenesis in the hippocampus. The learning and memory processes depend on this part of the brain. It is thought that Cotman *et al*. (2007), made a substantial addition to the field of neuroscience with their study by investigating the impact of physical activity on neurogenesis, the researchers used mouse models. According to their research, mice that participated in wheel running showed higher levels of neurogenesis. The human population has also been shown to exhibit this propensity. In a study by Herting & Nagel (2012), a group of adolescents engaged in a six-month aerobic exercise program reported significant enhanced their memory function and hippocampal volume.

The creation of neurotrophic factors, which are proteins that support the growth, survival, and functionality of neurons, has been linked to the cognitive benefits of aerobic exercise. The importance of Brain-derived neurotrophic factor (BDNF) comes from its role in cognitive function and synaptic plasticity. According to research (Pietrelli *et al*., 2018), aerobic exercise has the power to increase brain-derived neurotrophic factor (BDNF) levels in both human and animal participants. Increased levels of brain-derived neurotrophic factor (BDNF) have been discovered to be connected with better learning and memory.

The impact of neuroplasticity on changes in synapse structure. The basis for cognitive processes connected to learning and memory is neuroplasticity, the brain's amazing capacity to remodel and adapt in response to inputs. Since exercise encourages changes in the strength of connections between synapses, which ultimately result in gains in cognitive capacities, its influence on neuroplasticity is being acknowledged more and more. The research done in this area has shed light on the processes through which exercise-induced synaptic changes improve learning and memory-related cognitive skills.

Exercise's ability to enhance synaptic plasticity has been well demonstrated in numerous studies. This phenomenon describes the basic process by which connections between neurons are made and strengthened. The hippocampus and prefrontal cortex, two critical areas for the processes of learning and memory, have shown increased synaptic plasticity in response to aerobic exercise. Previous studies by Ferrer-Uris *et al*. (2022), have shown that exercise-exposed animals, specifically rats, showed greater synaptic plasticity. It has been discovered that improved performance on tests of spatial memory correlates with an increase in synaptic plasticity.

A key mechanism for improving information processing and memory formation is the strengthening of brain connections through exercise. It has been demonstrated that regular physical activity increases synapse efficiency, permitting improved neuronal communication. Elevated post-synaptic potentials and increased neurotransmitter release, which ultimately lead to improved neuronal signaling, are indicators of this occurrence.

Exercise causes synaptic alterations that affect neurotransmitter release and receptor density through molecular and cellular processes. Exercise has the potential to have an effect on neurotransmitter production, particularly glutamate, which is crucial for regulating synaptic plasticity. According to Lu *et al*. (2011), research, physical activity increases glutamate release, which aids in synapse strengthening.

Additionally, physical activity may have an impact on the receptor density on the post-synaptic membrane. For instance, animal studies by Ferrer-Uris *et al*. (2022), showed a beneficial relationship between exercise and the overexpression of NMDA receptors, which are essential for synaptic plasticity. The potency of synaptic connections is strengthened due to the elevation of receptor expression, which increases the sensitivity of neurons to neurotransmitters.

The processes of learning and memory are closely related to the phenomena of long-term potentiation (LTP), which is defined by the strengthening of synaptic connections through repeated stimulation. Exercise can increase synaptic plasticity, which in turn can boost long-term potentiation (LTP), aiding the brain's ability to encode and maintain information. Lynch's (2004) work offers as an example of how physical activity-induced improvements in long-term potentiation (LTP) might result in improved spatial learning and memory in mice.

## MATERIAL AND METHOD

**Animal Model and Groups.** 60 male C57BL/6 mice in the 8–10 week age range were brought in for the study. The animals were between 150 and 180 g in weight and kept in

the Central Animal Facility under standard laboratory the preferences, with a temperature of 21 ± 2 °C and a light cycle between 6:00 and 18:00 h. The Ethics Committee for animal Research gave its approval to the study's protocol. The Mice were randomly assigned to two groups: the exercise group (EGn=30) and the sedentary control group (SCn=30). The control group remained sedentary throughout the research, while the intervention group followed a long-term aerobic exercise program.

**Exercise Protocol. Treadmill Exercise Regimen:** The aerobic exercise program used by the exercise group was performed on a treadmill. This dynamic intervention took place over the course of 8 weeks, with 5 sessions per week. Each session lasted 30 minutes and consisted of a 5-minute warm-up at 40 % of maximum speed, a 20-minute exercise period at 60 % of maximum speed, and a 5-minute cool-down period. Weekly increases in exercise intensity, such as a 5 % weekly increase in speed, were used to control the regimen's progressive character.

**Cognitive and Learning Assessment**

**Morris water maze (MWM).** To measure cognitive skills related to spatial learning and memory, the Morris water maze (MWM) test was applied. The experimental set-up consisted of a 150 cm diameter circular pool that was filled with opaque water and maintained at room temperature. Within a certain region of the pool, a covert escape platform was placed.

**Acquisition phase:** Throughout the acquisition phase, the mice were put through a series of tests over the course of five consecutive days. Every mouse took part in four trials each day. In order to conduct the experiment, mice were dropped into the water and initially positioned facing one of the four directions—North, South, East, or West. Every trial's escape latency, which is the amount of time needed to get to the platform, was recorded. A mouse was then pointed in the direction of the platform if it couldn't find it after searching for 60 seconds. The mouse was allowed to rest on the platform for 15 seconds after each trial. In order to assess the learning curve, the average escape latencies for each day were determined.

**Probe Trial Period:** On the sixth day, a probing task was conducted to gauge spatial memory. The platform was taken out of the water. Every single mouse was placed into the pool from a different starting location and allowed 60 seconds of unfettered movement. The amount of time spent in the target quadrant, where the previous platform was located, as well as the number of crossings over the platform, were recorded.

**Test for Novel Object Recognition**

In the study of cognitive neuroscience, the Novel Object Recognition Test (NORT) is a popular experimental paradigm. It is intended to evaluate a person's capacity for object recognition and memory. Presenting is a test requirement. The novel object recognition test has two independent steps for evaluating recognition memory: habituation and testing.

**Habituation:** Each mouse was separately placed in an open-field arena with two identical objects, and they were free to explore the space for a total of 10 minutes. The mice were given the chance to become accustomed to the items and the arena during this phase.

**Testing:** The mice were reintroduced into the arena an hour after the habituation process had ended. During the habituation phase, a well-known object was changed out for a brand-new one. The mice had unrestricted access to the items for five minutes. The act of exploration was operationally defined as the inspection of an object through touch, smell, or vision while remaining less than 2 centimeters away. The amount of time spent studying each particular thing was recorded. The formula DI = (Time devoted to the novel object - Time allocated to the known object) / (Total duration of exploration) was used to calculate the discriminating index (DI).

**Tissues Collection and Analysis**

**The Brain Tissue Collection.** In order to assess the impact of persistent aerobic exercise on the production of new neurons and the capacity of synapses to modify and adapt, brain tissue was collected following an 8-week exercise program. The detailed methodology that follows outlines the steps used in order to collect brain tissue from the experimental mouse subjects.

**Euthanasia:** Following the workout, a group of mice was picked so that brain tissue could be collected from them. The mice were transferred to a chamber designed for euthanasia, where isoflurane gas was used to induce anesthesia in them. The lack of a withdrawal response and the absence of a respiratory rate response both indicated the existence of anesthesia.

**Perfusion:** Transcardial perfusion was carried out using physiological saline, then a solution consisting of 4 % paraformaldehyde in phosphate-buffered saline (PBS), to improve tissue fixation. A small surgical incision was made in the left ventricle of the heart, and a perfusion needle was then inserted to let solutions travel more easily through the circulatory system.

The first procedure included perfusing saline solution into the arteries to eliminate blood, and then the fixative (4 % paraformaldehyde in PBS) was applied to preserve the tissue's integrity.

**The Procedure of Brain Extraction.** Precision scissors were used to make a longitudinal incision along the midline of the scalp following the delivery of euthanasia and verification of anesthesia.

Precision scissors were used during a surgical procedure to make a midline cut in the skull, taking care to avoid damaging the brain tissue beneath.

The cerebral organ, which was kept moist by the application of cooled phosphate-buffered saline (PBS), was carefully exposed by carefully retracting the cranial flaps.

Using a spatula and blunt forceps, the brain was carefully removed from the cerebral cavity starting at the caudal limb.

The remaining cranial nerves and arteries were delicately severed in order to guarantee total evacuation of the brain.

The brain was meticulously dissected along the midline to separate the left and right hemispheres when it was necessary to conduct certain experiments that required hemisphere separation. (Fig. 1)

**The Preservation of Tissues.** In order to obtain a sufficient fixation, the brain tissues designated for histological investigation were placed in a fixative solution (4 % paraformaldehyde solution) permit additional histological analysis and held at a temperature of 4°C for a period of about 24 hours. Following fixation, the tissues were transferred to a cryoprotectant solution made of 30 % sucrose in PBS and stored there until further operations could be performed.

**Histological Analysis of Neurogenesis.** To evaluate neurogenesis, the brains were preserved in paraformaldehyde and subsequently sliced into 40 mm coronal sections using a vibratome. A systematic approach was employed to collect every sixth slice in order to guarantee an unbiased sampling process. The sections underwent immunohistochemical processing using an anti-doublecortin (DCX) antibody, which serves as a marker for immature neurons.

**Immunohistochemistry.** The sections were subjected to treatment with a blocking solution, such as 3 % normal goat serum, followed by incubation with the primary antibody targeting DCX at a dilution of 1:500. This incubation was carried out overnight at a temperature of 4°C. After the incubation of the primary antibody, the sections underwent subsequent incubation with a biotinylated secondary antibody, followed by an incubation with an avidin-biotin-peroxidase complex. The chromogen solution, such as 3,3'-diaminobenzidine (DAB), was employed to visualize the signal, leading to the formation of a brown precipitate in areas containing cells positive for DCX. The specimens were affixed onto slides, subjected to dehydration, and then covered with a coverslip for microscopic examination.

**Quantification.** The enumeration of DCX-positive cells was conducted inside certain regions of interest, such as the dentate gyrus of the hippocampus, employing a light microscope. Cell counts were standardized by dividing them by the area of the region, resulting in cell density values. The mean cell density per region was computed and subsequently employed for statistical analysis.

**Molecular Analysis.** The process of molecular analysis was conducted to investigate synaptic plasticity. Specifically, the hippocampal and prefrontal cortex tissues were subjected to homogenization in a lysis buffer. Subsequently, the protein concentrations in the homogenized samples were evaluated using a protein assay kit. A Western blot analysis was performed to evaluate the expression levels of synaptic markers, specifically synaptophysin and PSD-95.

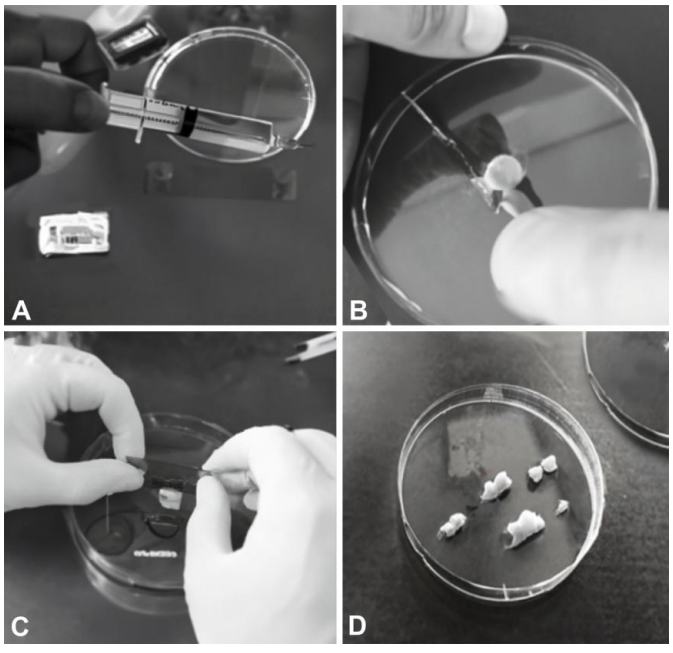


Fig. 1. The procedure for dissecting the mouse brain tissue block, depicting the required tools for preparation and the dorsal dissection process.

The Western blot technique, also known as protein immunoblotting, is a widely used laboratory method in molecular biology and biochemistry.

The protein samples underwent separation by the utilization of SDS-PAGE and were afterwards transferred onto nitrocellulose membranes. The membranes were obstructed and afterwards subjected to primary antibody incubation against synaptophysin (1:1000) and PSD-95 (1:1000) for an extended period at a temperature of 4°C. The membranes were subjected to incubation with secondary antibodies that were appropriately coupled to horseradish peroxidase. The visualization of protein bands was achieved by employing chemiluminescence reagents, followed by imaging utilizing a gel documentation system. The quantification of band intensities was performed utilizing image analysis tools.

## RESULTS

**Morris water maze test.** Over the course of the 5-day training session, the experimental group continuously displays a decrease in escape latency, indicating a greater rate of learning when compared to the control group ($F(1,58) = 11.78$, $p < 0.001$). Exercise-group participants showed a faster rate of platform position learning. The exercise group showed a statistically significant increase in the amount of time spent in the target quadrant when compared to the control group during the sixth day of the probing experiment ($t(58) = 4.02$, $p < 0.05$). This result implies that the exercise group had improved spatial memory retention. Furthermore, it was discovered that the exercise group crossed platforms more frequently ($t(58) = 3.15$, $p < 0.01$), adding more proof to the idea that their spatial memory recall was enhanced (Fig. 2).

**Novel Object Recognition Test Results.** The exercise group displays a notably higher DI compared to the control group, demonstrating enhanced recognition memory for novel objects. In the novel object recognition test, the exercise group exhibited a notably higher discrimination index compared to the control group ($t(28) = 3.67$, $p < 0.01$). This result indicates that mice subjected to the long-term aerobic exercise regimen demonstrated enhanced recognition memory for novel objects compared to sedentary mice (Fig. 3).

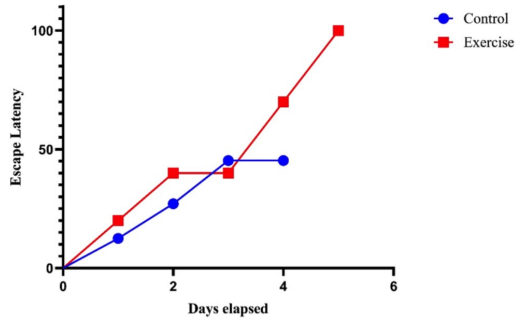


Fig. 2. The learning curve for both the exercise group and the control group during the Morris water maze test's acquisition phase. The vertical axis displays the escape latency, or how long it takes to find the platform. The number of days that the trials were run is depicted on the horizontal axis.

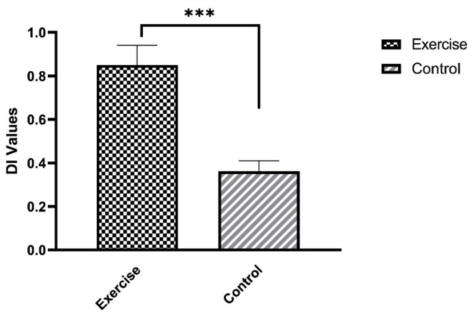


Fig. 3. This figure presents the discrimination index (DI) values for the exercise and control groups in the novel object recognition test. The y-axis represents the DI, and the x-axis represents the experimental groups.

**Neurogenesis Analysis.** The exercise cohort has a higher concentration of DCX-positive cells in the exercise cohort, $t(28) = 5.71$, $p < 0.001$). This result strongly implies that sustained aerobic exercise encourages neurogenesis in the hippocampus, a brain area linked to learning and memory (Fig. 4).

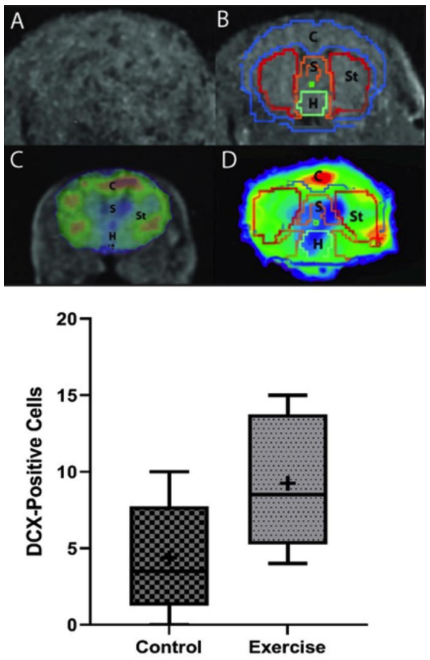


Fig. 4. Slices of the hippocampus are shown in representative images after being stained for doublecortin (DCX) immunohistochemistry. Panel A and C shows a section with a control group, while Panel B and D shows a segment with an exercise group.

**Molecular Analysis -Synaptic Plasticity.** When compared to the control group, the exercise group has higher levels of both synaptic markers ($t(28) = 3.86, p < 0.01$) and PSD-95 ($t(28) = 3.23, p < 0.05$) this consistence indicate aerobic exercise program effect on increased synaptic plasticity, which may have contributed to the observed cognitive improvements (Fig. 5).

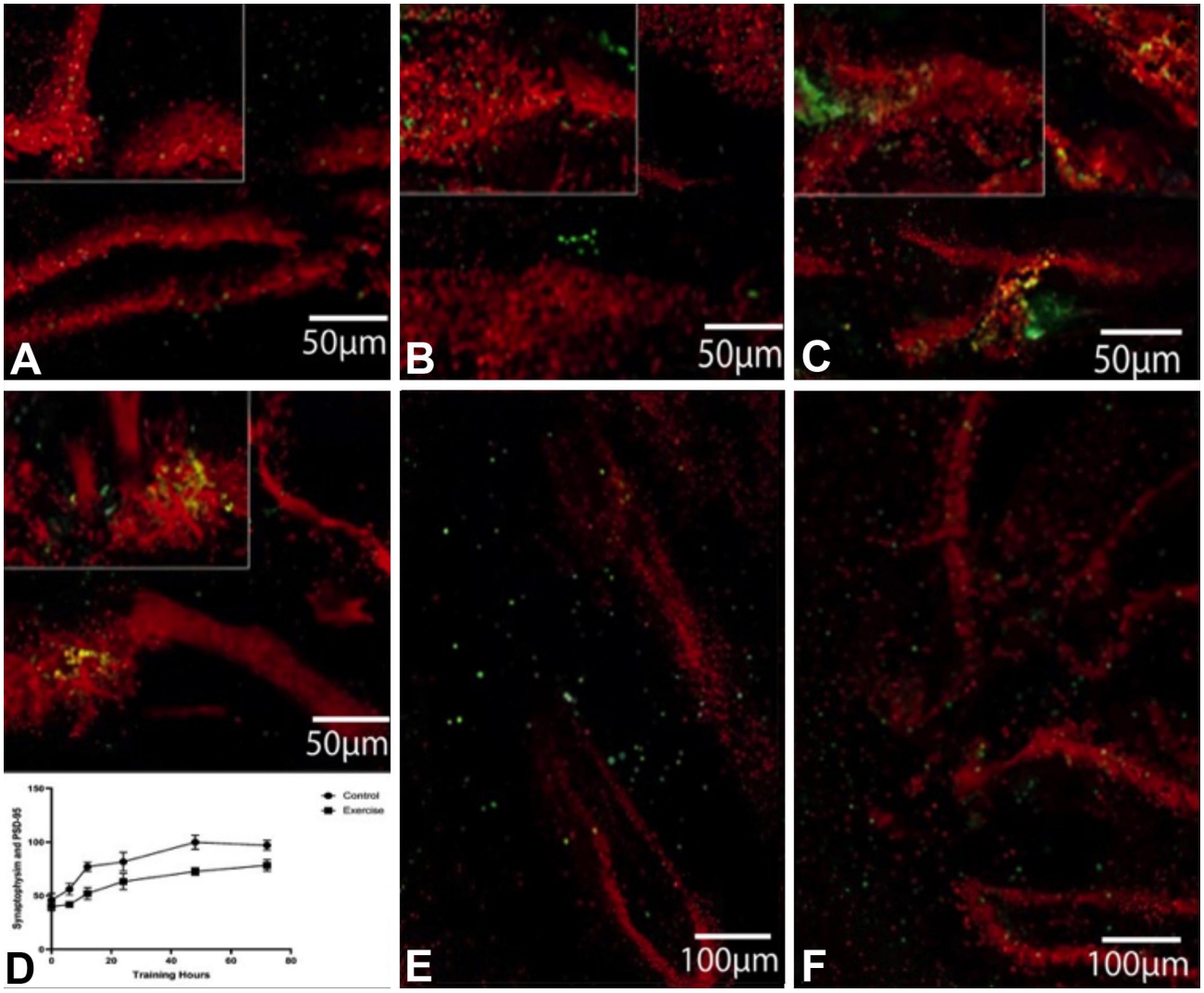


Fig. 5. The levels of synaptophysin and PSD-95 in hippocampus tissue samples from the exercise and control groups are shown in this figure.

## DISCUSSION

**Cognitive Improvements and Learning Enhancement.** The current study investigated the effects of a prolonged aerobic exercise program on synaptic plasticity, neurogenesis, and cognitive performance in mice. The findings suggest that persistent exercise over an extended period has the potential to significantly improve cognitive function, promote development of new neurons, and increase the plasticity of synaptic connections.

This study's conclusions are based in large part on the observed improvements in spatial learning and memory ability, which highlight the significant cognitive advantages of sustained exercise. The exercise group showed a continuous reduction in escape latency over the course of the 8-week training session in the Morris water maze test. Given that mice in the exercise group learned the spatial cues required to find the hidden escape platform more quickly, this reduction

denotes an increased capacity for learning and memory consolidation. The dramatic improvement in time spent in the target quadrant during the probing experiment not only shows increased memory recall but also suggests that the exercise group has developed a strong spatial cognitive map. Together, the reduced escape latency caused by exercise and the extended presence in the target quadrant suggest a complex cognitive improvement that goes beyond simple memory recall. These results are consistent with earlier studies that have shown how exercise might improve spatial learning tasks through mechanisms including not only memory but also sophisticated navigational techniques (Ang *et al*., 2006; Alomari *et al*., 2013).

Additionally, the exercise group's improved performance in the novel object identification test demonstrates the broad impact of exercise on cognitive areas. Associative learning is made easier because every day cognitive processes are supported by recognition memory, a crucial component of cognitive function. This cognitive skill is cooperatively orchestrated by the hippocampus and prefrontal cortex. The idea that long-term aerobic exercise can promote the sharpening of recognition memory through potential improvements in synaptic connections within these neural hubs is further supported by the agreement of the present study's findings with earlier research (Ahmadiasl *et al*., 2003).

However, cognitive advantages go beyond merely improving memory and spatial awareness. The intricate interaction between molecular foundations and brain circuitry highlights the variety of cognitive benefits of exercise. The underlying alterations involve a range of neurobiological adaptations that collectively strengthen the brain's ability for learning, memory, and cognitive resilience, even though the immediate consequences may show up as higher performance on certain cognitive tasks. Long-term aerobic exercise promotes cognitive gains through a dynamic tapestry of brain plasticity, neurotransmitter dynamics, and structural alterations.

These results support the idea that the brain is extraordinarily elastic and capable of adjusting to the environment in ways that go beyond the usual limitations of age and situation. The capacity of therapies like exercise to harness the brain's innate abilities and improve cognitive performance is highlighted by this malleability, or neuroplasticity. It is crucial to understand the underlying mechanisms at the molecular, cellular, and network levels in order to realize the full potential of cognitive advances.

**Neurogenesis and Cognitive Plasticity.** A key factor in cognitive plasticity—the brain's ability to adapt, learn, and rewire itself—is neurogenesis, a complicated process that creates new neurons from neural stem cells. Long-term aerobic activity appears to be a facilitator in this cognitive enhancing process, and the fundamental ramifications of neurogenesis go far beyond being just a mechanism for cell replacement. The considerable increase in DCX-positive cell density observed in the hippocampus dentate gyrus of the exercise group in the current study sheds information on the major influence of exercise on neurogenesis. This is in line with other research (Gomes da Silva & Arida, 2015) that showed exercise has the capacity to induce hippocampus regeneration, creating a neuronal environment that is favorable for cognitive improvement.

Exercise-induced neurogenesis appears as a dynamic stage in the hippocampus, a region well-known for its critical function in spatial learning and memory. According to Ma *et al*. (2017), the hippocampus has a special plasticity that plays a crucial part in the development of cognition. It is hypothesized that increased neurogenesis bestows a number of favorable neurobiological characteristics that collectively support cognitive performance.

The increase in neural network complexity is one such quality. The encoding and retrieval of complex information may be made easier by the expansion of brain connections caused by the integration of additional neurons into preexisting circuits (Ma *et al*., 2017). In addition to increasing information storage capacity, this complexity encourages the development of diverse brain patterns that are essential for complex cognitive processes like pattern recognition and associative learning. The higher density of DCX-positive cells in the exercise group shows that these new neurons are integrated into the hippocampus network, contributing to the dynamic structural changes linked to cognitive plasticity.

Additionally, neurogenesis brought on by exercise may improve the effectiveness of information processing. A dynamic ensemble of cells with distinct electrical characteristics is created by the inflow of new neurons, fostering a variety of functional contributions to cognitive processes. This variety gives the hippocampus access to a variety of computational resources, which improves its capacity to handle data across many cognitive domains (Shohayeb *et al*.,2018). A potential richness of this neuronal variety is suggested by the increased DCX-positive cell density seen in the workout group.

It is also suggested that exercise-induced neurogenesis can enhance adaptive cognitive abilities.

These abilities are crucial for an organism's survival. The hippocampus's adaptability, which is enhanced by the inflow of new neurons, allows for situational adaptability while enhancing the cognitive repertoire. Moreover, exercise-induced neurogenesis has a crucial role in the context of neurological diseases and cognitive aging. The sensitivity of the hippocampus structure to age-related deterioration and neurodegenerative diseases emphasizes the need for quick action to promote neurogenesis (Rodgers *et al*., 2013). As such, the foundation of cognitive plasticity is adaptability. This neurogenesis ability allows for learning, memory development, and the adaptive modification and response to environmental and situational factors. The results of the current study support the idea that regular aerobic exercise, especially in groups at risk for impaired neuroplasticity, may act as a preventive or mitigating measure against cognitive decline.

**Cognitive Benefits of Synaptic Plasticity.** The complicated network of cognitive processes has its roots in the captivating dance of synapses, which is supported by synaptic plasticity as a neural foundation. Learning and memory are cellular processes that depend on synapses' amazing capacity to become stronger or weaker in response to brain activity. Our ability to learn new information, access previously stored memories, and adjust to unfamiliar events is supported by the symphony of cognitive transformations that are orchestrated by these dynamic alterations inside the synapses.

**Aerobic Exercise and Improved Synaptic Plasticity:** The elevated levels of the synaptic markers synaptophysin and PSD-95 in the exercise group's hippocampal tissue support the idea that synaptic plasticity is improved by regular aerobic exercise. These indicators of synapse density and neural connection tell the tale of how the brain adapts to continuous physical exertion. This story fits with earlier research that showed how exercise can increase synaptic markers, shaping neuronal networks that are best suited for cognitive performance (Lu *et al*., 2011; Shohayeb *et al*., 2018).

**Functional adaptations and structural changes:** Prefrontal cortex and the hippocampus, esteemed areas important for memory, learning, and higher-order cognitive processes, emerge as the sites of exercise-induced structural and functional change. The dynamic alteration that results from the tapestry of changes, which range from increased spine density to extended dendritic branching, is a reflection of the brain's unceasing adaptation to cognitive demands. The orchestra of structural improvements is further accentuated by the exercise-driven increase in neurotransmitter release (Pietrelli *et al*., 2018).

**Potential Mechanisms Underpinning Cognitive Benefits:** Long-term aerobic exercise has the potential to choreograph a symphony of structural alterations that will lead to the orchestral accuracy of cognitive processes. The expanded dendritic arborizations provide a larger canvas on which to paint neural information, while the increased synaptic density creates a favorable environment for strong neural connections (Ferrer-Uris *et al*., 2022). Exercise-induced increases in neurotransmitter release and increased spine density may boost the effectiveness of neural transmission, ensuring that cognitive instructions travel across neural circuits with unrivaled speed.

## CONCLUSION

In conclusion, our investigation offers a useful vantage point from which to assess the effects of continuous aerobic exercise on murine mice' cognitive faculties. A recognizable theme refrain that resonates throughout the fields of spatial learning, recognition memory, neurogenesis, and synaptic plasticity is shown in our findings. This synthesis clarifies the role of physical activity as a significant modulator of cognitive performance and neural plasticity, supporting the possibility of a synergistic relationship between physical activity and the cognitive domain. However, these discoveries are supported by a deeper orchestration, prompting the need for more research to identify the precise underlying mechanisms and cross the disciplinary boundaries where physical and cognitive abilities interact.

The results of the study demonstrate a significant enhancement of spatial learning ability, as seen by a reduction in escape latency during the Morris water maze test. The findings suggest increased spatial cognitive ability brought on by the workout program. Coherently, the exercise group's improved performance in the novel object recognition test emphasizes cognitive augmentation, underscoring the complex intricacies of recognition memory. This concordance emphasizes the multifaceted and all-encompassing nature of cognitive improvements brought about by sustained physical involvement.

The information on neurogenesis is a cornerstone of the discussion of cognitive plasticity. Exercise's role as a catalyst for promoting neurogenesis is empirically supported by the apparent increase in DCX-positive cell density within the hippocampus dentate gyrus. The idea that exercise-related influence manifests beyond cognitive domains and catalyzes intricate alterations within the brain architecture itself is postulated as a result of the convergence of structural enhancements and enhanced neural signaling.

### Implications for Future studies

a) The results of this study have significance for both clinical and fundamental applications of neuroscience. The rising corpus of research demonstrating the efficacy of non-pharmacological therapies for cognitive health benefits from an understanding of the mechanisms underlying the cognitive advantages of aerobic exercise. The application of these discoveries to people may help develop methods to reduce cognitive deterioration brought on by aging and neurodegenerative diseases.

b) Future studies should focus on the molecular mechanisms through which exercise affects neurogenesis and synaptic plasticity. Deeper understanding of the mechanisms underlying cognitive gains can be gained by examining the involvement of neurotrophic factors, such as brain-derived neurotrophic factor (BDNF), and how they interact with processes brought on by exercise. Additionally, investigating how various exercise intensities, durations, and timings affect cognitive outcomes might direct the creation of tailored exercise therapies.

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**RESUMEN:** Tanto el mundo académico como el popular han prestado mucha atención al vínculo entre el ejercicio y el rendimiento cognitivo. Es cada vez más importante comprender los numerosos mecanismos por los cuales el ejercicio podría influir en las capacidades cognitivas en vista de los continuos problemas sociales causados por el envejecimiento de la población y la prevalencia de trastornos asociados al deterioro cognitivo. Una cantidad cada vez mayor de evidencia que muestra una asociación favorable entre la actividad física y el bienestar cognitivo sirve como base para justificar el estudio de los efectos del ejercicio sobre la función cognitiva y la capacidad de aprendizaje. El estudio se realizó en ratones machos adultos C57BL/6 utilizándose en los ejercicios una cinta rodante durante 8 semanas. El grupo de ejercicio realizó 5 sesiones por semana aumentando gradualmente la intensidad del protocolo en un 5 % cada semana. Las evaluaciones cognitivas de los ratones se realizaron utilizando las pruebas Morris Water Maze y Novel Object Recognition. El impacto a largo plazo en la capacidad de aprendizaje se evaluó mediante inmunohistoquímica y análisis molecular de los tejidos del hipocampo y la corteza prefrontal de los tejidos cerebrales de los animales. Los hallazgos mostraron mejoras en las habilidades de aprendizaje espacial, la memoria de reconocimiento y una mayor plasticidad sináptica indicada por unos creadores sinápticos elevados. El estudio subraya el papel del ejercicio aeróbico a largo plazo para aumentar el rendimiento cognitivo. No sólo contribuye a la comprensión de la interacción entre la neuroplasticidad y los beneficios cognitivos, sino también al creciente conjunto de investigaciones sobre el impacto del ejercicio en la función cognitiva.

**PALABRAS CLAVE: Ejercicio aeróbico de larga duración; Función cognitiva; Capacidad de Aprendizaje; Neuroplasticidad; Aprendizaje espacial.**

## REFERENCES

Ahmadiasl, N.; Alaei, H. & Hänninen, O. Effect of exercise on learning, memory and levels of epinephrine in rats' hippocampus. *J. Sports Sci. Med., 2(3)*:106-9, 2003.

Alomari, M. A.; Khabour, O. F.; Alzoubi, K. H. & Alzubi, M. A. Forced and voluntary exercises equally improve spatial learning and memory and hippocampal BDNF levels. *Behav. Brain Res., 247*:34-9, 2013.

Ang, E. T.; Dawe, G. S.; Wong, P. T. H.; Moochhala, S. & Ng, Y. K. Alterations in spatial learning and memory after forced exercise. *Brain Res., 1113(1)*:186-93, 2006.

Cotman, C. W.; Berchtold, N. C. & Christie, L. A. Exercise builds brain health: key roles of growth factor cascades and inflammation. *Trends Neurosci., 30(9)*:464-72, 2007

Ferrer-Uris, B.; Ramos, M. A.; Busquets, A. & Angulo-Barroso, R. Can exercise shape your brain? A review of aerobic exercise effects on cognitive function and neuro-physiological underpinning mechanisms. *AIMS Neurosci., 9(2)*:150-74, 2022.

Gomes da Silva, S. & Arida, R. M. Physical activity and brain development. *Expert Rev. Neurother., 15(9)*:1041-51, 2015.

Herting, M. M. & Nagel, B. J. Aerobic fitness relates to learning on a virtual Morris Water Task and hippocampal volume in adolescents. *Behav. Brain Res., 233(2)*:517-25, 2012.

Ikeda, T.; Mishima, K.; Aoo, N.; Harada, K.; Liu, A. X.; Egashira, N.; Iwasaki, K.; Fujiwara, M. & Ikenoue, T. Rehabilitative training tasks improve spatial learning impairment in the water maze following hypoxic-ischemic insult in neonatal rats. *Pediatr. Res., 59(1)*:61-5, 2006.

Liu, H. L.; Zhao, G.; Cai, K.; Zhao, H. H. & Shi, L. D. Treadmill exercise prevents decline in spatial learning and memory in APP/PS1 transgenic mice through improvement of hippocampal long-term potentiation. *Behav. Brain Res., 218(2)*:308-14, 2011

Lu, B.; Nagappan, G. & Lu, Y. BDNF and synaptic plasticity, cognitive function, and dysfunction. Handb. Exp. Pharmacol., 220:223-50, 2014.

Ma, C. L.; Ma, X. T.; Wang, J. J.; Liu, H.; Chen, Y. F. & Yang, Y. Physical exercise induces hippocampal neurogenesis and prevents cognitive decline. *Behav. Brain Res., 317*:332-9, 2017.

Pietrelli, A.; Matkovic, L.; Vacotto, M.; Lopez-Costa, J. J.; Basso, N. & Brusco, A. Aerobic exercise upregulates the BDNF-Serotonin systems and improves the cognitive function in rats. *Neurobiol. Learn. Mem., 155*:528-42, 2018.

Rodgers, S. P.; Trevino, M.; Zawaski, J. A.; Gaber, M. W. & Leasure, J. L. Neurogenesis, exercise, and cognitive late effects of pediatric radiotherapy. *Neural Plast., 2013*:698528, 2013.

Shohayeb, B.; Diab, M.; Ahmed, M. & Ng, D. C. H. Factors that influence adult neurogenesis as potential therapy. *Transl. Neurodegener., 7*:4, 2018.

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