



The Effect of Endurance Training on Some Angiogenic Markers in Brain Tissue in an Experimental Autoimmune Encephalomyelitis Model

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ARTICLE INFO	ABSTRACT
<p><i>Article type:</i> Research Paper</p>	<p>Introduction: Experimental autoimmune encephalomyelitis (EAE), a model of multiple sclerosis, involves neuroinflammation and cerebrovascular dysfunction. However, the impact of exercise on cerebral angiogenic and vasoactive mediators remains unclear. Therefore, the present study aimed to investigate the effects of a five-week endurance training (ET) protocol on the gene expression levels of vascular endothelial growth factor (VEGF) and endothelin-1 (ET-1) in brain tissue in an experimental model of EAE.</p> <p>Methods: In this experimental study, fourteen female Sprague-Dawley rats with EAE (induced using complete Freund's adjuvant) were randomly assigned to either the EAE (n=7) or EAE+ET (n=7) groups. To assess baseline differences, seven healthy rats were included as a healthy control group (HC). Endurance training started ten days after EAE induction. Rats underwent one week of treadmill familiarization (6 m/min, 11% incline, 5–25 min/day), followed by five weeks of moderate-intensity running (11 m/min, 30 min/day, 5 sessions/week). Data were analyzed using one-way ANOVA followed by Tukey's post hoc test (P ≤ 0.05).</p> <p>Results: The gene expression levels of VEGF and ET-1 were significantly higher in the EAE group compared to the HC group (P = 0.001). However, both VEGF and ET-1 expression were markedly reduced in the EAE+ET group relative to the EAE group (P ≤ 0.05).</p> <p>Conclusion: The findings suggest that endurance exercise may exert beneficial effects by attenuating inflammatory responses and angiogenic activity in brain tissue following EAE induction. Nonetheless, given the limited evidence on how exercise modulates VEGF and ET-1 expression in the brain under autoimmune conditions, further research is warranted.</p>
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Introduction

Multiple sclerosis (MS) is a chronic inflammatory disease of the central nervous system characterized by immune-mediated demyelination and neurodegeneration. Epidemiological data indicate that the global prevalence of MS has increased substantially; in recent years, approximately 2.8 million individuals worldwide have been affected by the disease. According to 2019 estimates, the prevalence of MS ranges from 30.5 to 89 cases per 100,000 population, with an ongoing upward trajectory (1). From a pathophysiological perspective, immune dysregulation, impaired cerebral blood flow, and disturbances in

neurotrophin signaling within the central nervous system (CNS) collectively contribute to neuronal injury and structural brain damage. However, due to the inherent limitations of direct investigations on human CNS tissue, researchers often utilize animal models—most notably experimental autoimmune encephalomyelitis (EAE)—to elucidate underlying disease mechanisms (2).

It is well established that inflammatory dysregulation in MS is associated with cerebrovascular abnormalities, including alterations in vascular structure and permeability, dysregulation of iron metabolism, and collagen deposition—particularly type IV

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collagen (Col-IV)—as well as enhanced vascular fibrinolysis (3). Within the CNS, vascular endothelial growth factor (VEGF) expression is upregulated in response to inflammatory stimuli, where it functions as a proinflammatory mediator. This upregulation is thought to enhance cerebral blood flow and facilitate the dissemination of inflammatory mediators to other brain regions (4). Moreover, elevated VEGF expression in EAE models has been reported to stimulate Toll-like receptor 2 and 4 (TLR2/4) signaling, thereby inducing endothelin-1 (ET-1) expression. This downstream cascade subsequently promotes the release of tumor necrosis factor- α (TNF- α) and interleukin-1 (IL-1), ultimately leading to vascular injury and demyelination within the brain (5).

Furthermore, numerous studies have emphasized the importance of identifying effective and noninvasive strategies for the prevention and management of chronic diseases as a central priority in biomedical research (6). Among these strategies, regular physical activity has consistently been recognized as an effective approach for improving metabolic and other chronic conditions (7). Evidence indicates that regular exercise exerts favorable effects in patients with MS as well as in animal models of EAE by enhancing quality of life, improving physical and cognitive performance, reducing chronic inflammatory risk factors, and modulating immune and inflammatory responses (8). In a recent study, aerobic exercise improved walking performance and increased markers of myelin repair in cuprizone-exposed mice (9). Similarly, high-intensity interval training (HIIT) has been reported to attenuate neuroinflammation, regulate immune system function, and inhibit the progression of EAE-induced neurological disturbances (10). Although evidence regarding VEGF alterations in the brain under EAE conditions remains limited, previous findings have shown that continuous high-intensity exercise decreased inflammatory markers such as very late antigen-4 (VLA-4) and lymphocyte function-associated antigen-1 (LFA-1), reduced cerebral blood flow, and downregulated intercellular adhesion molecule (ICAM) and vascular cell adhesion molecule (VCAM) expression in cerebral vessels of EAE rats (11). Furthermore, a clinical trial reported that six weeks of regular exercise reduced serum TNF- α and increased VEGF levels among

individuals with MS (12). In another experimental study, four weeks of aerobic exercise upregulated the gene expression of tight junction proteins occludin and claudin-5, both crucial for maintaining blood-brain barrier (BBB) integrity. This exercise-induced upregulation was associated with decreased BBB permeability in EAE-induced mice (13).

Despite the available evidence, while exercise is recognized as a promoter of angiogenesis, limited data exist regarding its effects on key angiogenic mediators such as vascular endothelial growth factor (VEGF) and endothelin-1 (ET-1) in the brain following EAE induction. Given the pivotal role of VEGF and ET-1 in cerebrovascular pathology, evaluating their gene expression in brain tissue under EAE conditions may offer mechanistic insight into exercise-induced neurovascular adaptations. This study uniquely investigates how aerobic exercise modulates VEGF and ET-1 in brain tissue of EAE rats, providing novel insights with potential implications for MS management. Therefore, the present study aimed to investigate the effects of a five-week endurance training (ET) protocol on the gene expression levels of VEGF and ET-1 in brain tissue in an experimental model of autoimmune encephalomyelitis (EAE).

Methods

Animal Care and Housing

This experimental, fundamental research was conducted on twenty-three female Sprague-Dawley rats aged 8–10 weeks and weighing 210 ± 10 g, obtained from the Laboratory Animal Breeding Center of Islamic Azad University, Marvdasht Branch. After transportation to the Animal Exercise Physiology Laboratory, the animals were acclimatized for seven days under controlled laboratory conditions. All experimental procedures were conducted in accordance with established ethical guidelines for animal research and the principles of the Helsinki Declaration. The study was approved by the Ethics Committee of Biomedical Research at Islamic Azad University, Khorasgan Branch, Isfahan, Iran, under the approval code IR.IAU.M.REC.1403.600. Throughout the study, rats were maintained under standard environmental conditions, including a temperature of 22–24 °C, relative humidity of 55–60%, and a 12-hour light/dark cycle. Animals were housed in polycarbonate cages with

sterilized wood-shaving bedding and had ad libitum access to standard laboratory chow and water. All efforts were made to minimize animal discomfort during injections and to adhere to standard protocols for tissue collection.

Induction of Experimental Autoimmune Encephalomyelitis (EAE)

One day after the acclimatization period, ten guinea pigs were obtained from the Pasteur Institute of Iran to prepare spinal cord homogenate for EAE induction. Guinea pig spinal cord tissue was specifically chosen as a neural homogenate to enhance the immunogenicity of the preparation, allowing for more efficient targeting of the rat central nervous system when combined with Complete Freund's Adjuvant (CFA), thereby facilitating reliable EAE induction. Following anesthesia with ketamine and xylazine, the spinal cords were excised, immediately frozen in liquid nitrogen, and pulverized under cryogenic conditions to obtain a fine powder. The homogenized spinal cord tissue was then mixed with an equal volume of normal saline and continuously stirred at 5 °C to form a uniform suspension. An equal volume of Complete Freund's Adjuvant (CFA) was subsequently added to the homogenate (1:1 ratio) and emulsified using a shaker to obtain a stable emulsion. Sixteen rats were anesthetized with ketamine and xylazine, then injected subcutaneously with 400 µL of the antigen-CFA emulsion into the hind limb and 100 µL into the footpad using a 25-gauge needle. Female rats were specifically selected for this study, as this sex is more susceptible to EAE induction, and previous studies investigating EAE similarly utilized female animals to ensure consistency and reproducibility. Following induction, animals were monitored daily and scored for neurological symptoms based on a standardized clinical scale: 0 = no symptoms; 1 = impaired tail movement; 2 = tail paralysis; 3 = gait disturbance; 4 = paralysis of one limb; 5 = paralysis of both hind limbs; 6 = tetraplegia; and 7 = death (14,15). Two rats died within eight days post-induction, and fourteen successfully developed EAE. These animals were categorized based on their clinical scores and randomly assigned to two groups: (1) EAE (control) and (2) EAE + ET (exercise-trained). Additionally, seven healthy rats were included as a healthy control (HC) group to establish baseline values and

assess the direct effects of disease induction on the study variables.

Endurance Training Protocol

Endurance training was initiated approximately 10 days after EAE induction. During the first week, rats were gradually acclimated to the treadmill by exercising daily at 6 m/min with an 11° incline for 5–25 minutes. The main training protocol was then carried out for five consecutive weeks, with rats running at a constant speed of 11 m/min. The duration of each session started at 25 minutes in the first week and was increased by 2 minutes each subsequent week, reaching 35 minutes by week five. This protocol was selected based on previous evidence demonstrating its neuroprotective effects in rodents with cognitive impairments, including models of Parkinson's disease and experimental autoimmune encephalomyelitis (14,15).

Tissue Dissection and Sampling

Forty-eight hours after the final training session—and following a 12-hour fasting period to eliminate the acute effects of exercise—all rats were anesthetized using a ketamine (80 mg/kg) and xylazine (15 mg/kg) mixture (Alfasan, Netherlands). After complete anesthesia, the cranial vault was carefully removed using surgical scissors and forceps. The brain tissue was gently dissected using fine scalpels (size 5) and immediately transferred into sterile microtubes for tissue preservation. Samples were rapidly immersed in liquid nitrogen for 20 minutes and subsequently stored at -70 °C until molecular analyses were performed.

Assessment of VEGF and ET-1 Gene Expression in Brain Tissue

Gene expression levels of *VEGF* and *ET-1* were determined using real-time polymerase chain reaction (RT-qPCR). Approximately 50 mg of frozen brain tissue was used for total RNA extraction following the manufacturer's protocol (Qiagen, Germany). RNA purity and integrity were verified through agarose gel electrophoresis and optical density assessment at 260 nm using a PicoDrop spectrophotometer (Sigma, USA). Complementary DNA (cDNA) was synthesized from purified RNA using the RevertAid cDNA Synthesis Kit (Thermo Fisher Scientific, K1621, USA) according to the manufacturer's instructions. Specific primers for *VEGF*, *ET-1*, and the internal reference gene

TATA-box binding protein (TBP) were designed using the NCBI Primer-BLAST tool and validated for specificity and efficiency (primer sequences are presented in Table 1). Quantitative gene expression was analyzed using a qReal-Time PCR

system. The relative expression of target genes was calculated using the $2^{-\Delta\Delta CT}$ method, with TBP serving as the endogenous control to normalize the data.

Table 1. Primer sequences used for real-time PCR analysis

Genes	Primer Sequences	Sizes (bp)
VEGF-A	Forward: 5'- ACTTGAGTTGGGAGGAGGATGTC -3'	183
	Reverse: 5'- GGATGGGTTTGTCTGTTTCTGG -3'	
Endothelin-1 (ET-1)	Forward: 5'- ACAAGAAGTCCGAGCCCAA -3'	245
	Reverse: 5'- CACGGGGCTCTGTAGTCAAT-3'	
TBP	Forward: 5'- GCGGGTTCATGAAATCCAGT-3'	147
	Reverse: 5'- AGTGATGTGGGGACAAAACGA -3'	

Statistical Analysis

Data normality was assessed using the Shapiro-Wilk test. One-way analysis of variance (ANOVA) was then performed to evaluate differences among groups. When significant differences were detected, Tukey's post hoc test was applied to determine pairwise group differences. All statistical analyses were conducted using SPSS software (version 26, IBM, USA), and a significance level of $P \leq 0.05$ was considered for all tests.

Results

One-way ANOVA revealed significant differences in *VEGF* ($F = 39.38$, $P = 0.001$) and *ET-1* ($F = 18.45$, $P = 0.001$) gene expression levels among the experimental groups.

Post hoc Tukey's test indicated that *VEGF* expression was significantly higher in the EAE group compared to the healthy control (HC) group ($P = 0.001$). In contrast, *VEGF* levels were significantly reduced in the EAE + ET group relative to the EAE group ($P = 0.001$), although they remained elevated compared to the HC group ($P = 0.006$) (Figure 1).

Similarly, *ET-1* expression was significantly elevated in the EAE group compared to HC ($P = 0.001$). Endurance training markedly decreased *ET-1* levels in the EAE + ET group relative to the EAE group ($P = 0.01$), yet these levels were still significantly higher than those in HC ($P = 0.001$) (Figure 2)

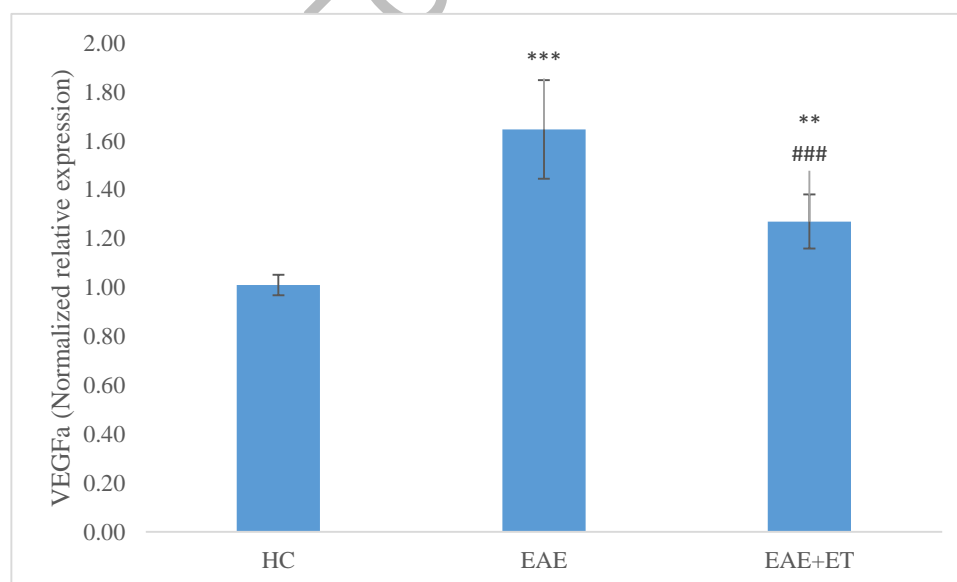


Figure 1. *VEGF* gene expression levels in brain tissue of rats across experimental groups.

HC: healthy control; EAE: experimental autoimmune encephalomyelitis control; EAE+ET EAE+ Endurance training. *** $P \leq 0.001$ and ** $P \leq 0.01$ vs. HC; ### $P \leq 0.001$ vs. EAE

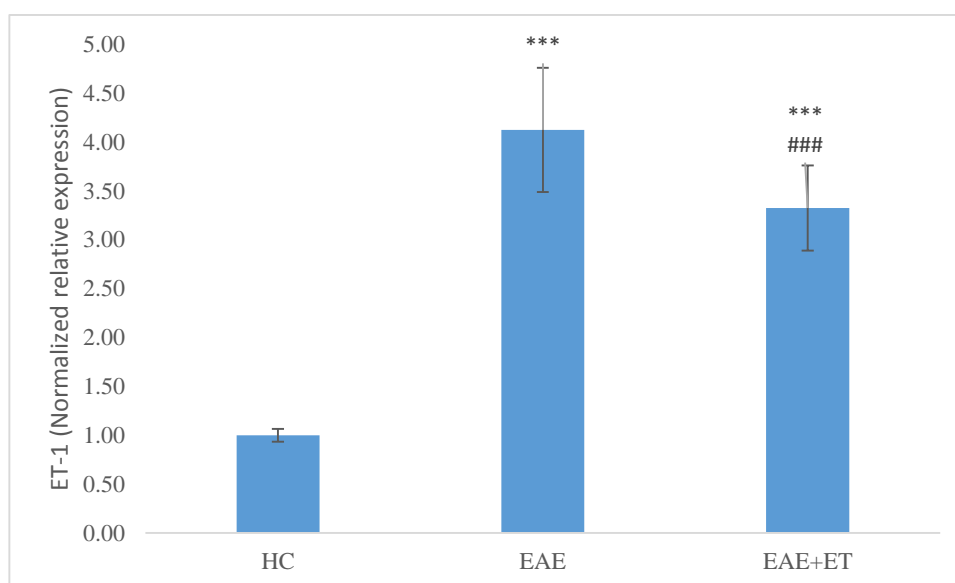


Figure 2. *ET-1* gene expression levels in brain tissue of rats across experimental groups.

HC: healthy control; EAE: experimental autoimmune encephalomyelitis control; EAE+ET EAE+ Endurance training. *** $P \leq 0.001$ vs. HC; ### $P \leq 0.001$ vs. EAE.

Discussion

The present study demonstrated that EAE induction significantly upregulated *VEGF* and *ET-1* gene expression in rat brain tissue. Mechanistically, it appears that the inflammatory cascade triggered by EAE initially stimulates the expression of proteins involved in the transcription of proinflammatory mediators. This signaling pathway is initiated following activation of Toll-like receptors (TLRs), such as TLR2, TLR3, and TLR4. Subsequently, cyclooxygenases in M1-type macrophages are activated, leading to the activation of nuclear factor kappa B (NF- κ B) and interferon-gamma, which ultimately results in the accumulation of inflammatory cytokines, including TNF- α , IL-1 β , and IL-6, in brain tissue. These proteins enhance the expression of adhesion molecules VCAM-1 and ICAM-1, which in turn increase ET-1 levels in cerebral vessels and promote vascular fibrillation and pathological remodeling (5). Physiologically, ET-1 contributes to vascular constriction and dilation, a process that is closely associated with nitric oxide (NO) activation. Furthermore, NO-mediated activation of nitric oxide synthase may enhance endothelial proliferation, regulate water and electrolyte balance, and promote VEGF expression, thereby facilitating angiogenesis in inflamed cerebral vessels (16). In this context, increased circulating

VEGF has been reported to support angiogenesis and cerebral perfusion, enhancing the accessibility of angiotensin II receptors and angiotensin-converting enzyme 2 (ACE2), which may exacerbate brain injury in SARS-CoV-2 infection (17). Conversely, VEGF and membrane-bound matrix metalloproteinase 9 (MMP9) serve as beneficial mediators in post-stroke rehabilitation, promoting vascular repair and functional recovery (18). Additionally, in Alzheimer's disease, elevated VCAM-1 and ICAM-1 expression in cerebral vessels is associated with reduced cerebral blood flow, whereas VEGF upregulation can counteract these effects and restore perfusion (19). These seemingly contradictory outcomes may reflect disease-specific mechanisms: in stroke and Alzheimer's disease, impaired blood flow contributes to pathology, whereas in autoimmune and inflammatory disorders, increased blood flow may exacerbate inflammatory injury to other tissues.

The results of the present study also demonstrated that ET reduced *VEGF* and *ET-1* expression in the brain tissue of EAE-induced rats. Although most studies report that exercise enhances angiogenesis, upregulates neurotrophins, and improves cognitive function, it is important to recognize that increased cerebral blood flow and angiogenesis may also

facilitate leukocyte infiltration and the migration of inflammatory mediators across brain regions (11). Conversely, the anti-inflammatory effects of exercise following EAE have been linked to enhanced M2 macrophage activity in brain tissue, associated with increased expression of IL-10, a key anti-inflammatory cytokine. Additionally, exercise may improve cerebral perfusion through reinforcement of the BBB. Both voluntary and forced exercise have been shown to upregulate tight junction proteins, such as occludin and claudin-2, thereby enhancing BBB integrity and restricting leukocyte migration into the CNS (13). Moreover, regular physical activity may downregulate NF- κ B signaling, increase neurotrophin expression, activate antioxidant transcription pathways, and inhibit cyclooxygenases, collectively contributing to reduced *ET-1* expression (20). While few studies have directly investigated the effects of exercise on *VEGF* and *ET-1* expression in brain tissue, one clinical trial reported that a single exercise session and a six-week exercise program significantly reduced serum TNF- α levels, whereas VEGF levels increased after the first and eighteenth sessions in patients with MS (12). In another study, high-intensity interval training was shown to decrease proteolipid transport to the CNS, reduce *VCAM-1* and *ICAM-1* expression, lower inflammatory cytokines and chemokines in brain tissue, and improve BBB function in an EAE model (11). Aerobic exercise has also been reported to reduce *ET-1* levels while improving vasodilation in postmenopausal women (21). The findings of the present study align with these observations, suggesting that the anti-inflammatory effects of aerobic exercise and enhanced neurotrophin activity may underlie the observed reductions in *ET-1* expression. It is noteworthy that reductions in serum inflammatory mediators and increased circulating VEGF primarily reflect peripheral responses and may not fully capture molecular changes within cerebral tissue, limiting interpretation of central effects. Consequently, differences in measurement sites may partly account for inconsistencies among previous studies. Given the limited existing data, the scarcity of investigations examining exercise-induced effects on VEGF and *ET-1* in brain tissue following EAE represents a substantial knowledge gap and highlights the novelty of the present study. Moreover, the study did not

include direct assessments of blood-brain barrier (BBB) function, which is critical for understanding cerebrovascular adaptations to exercise. Future research should therefore evaluate BBB integrity and permeability alongside key inflammatory mediators, including TNF- α , IL-1 β , and IL-6, to provide a more comprehensive understanding of exercise-induced neurovascular adaptations.

Conclusion

Based on the findings of the present study, endurance exercise appears to exert beneficial effects by reducing inflammatory mediators and modulating angiogenic factors in the brain following EAE induction. However, given the limited available evidence on the effects of exercise on VEGF and *ET-1* expression in brain tissue post-EAE, further studies are warranted to elucidate the underlying molecular mechanisms.

References

1. Moghtaderi A, Shahidi-Pourakbari M, Izadi S, Khosravi A, Hashemzahi Z. Ongoing increase in incidence and prevalence of multiple sclerosis in south-eastern Iran: A three decade study. *Mult Scler Relat Disord*. 2023;71:104557.
2. Calvillo-Robledo A, Ramírez-Farías C, Valdez-Urias F, Huerta-Carreón EP, Quintanar-Stephano A. Arginine vasopressin hormone receptor antagonists in experimental autoimmune encephalomyelitis rodent models: A new approach for human multiple sclerosis treatment. *Front Neurosci*. 2023;17:1138627.
3. J Karlik S, A Roscoe W, Patinote C, Contino-Pepin C. Targeting vascular changes in lesions in multiple sclerosis and experimental autoimmune encephalomyelitis. *Cent Nerv Syst Agents Med Chem (Formerly Curr Med Chem Nerv Syst Agents)*. 2012;12(1):7-14.
4. Girolamo F, Coppola C, Ribatti D, Trojano M. Angiogenesis in multiple sclerosis and experimental autoimmune encephalomyelitis. *Acta Neuropathol Commun*. 2014;2(1):84.
5. Jin Y-H, Kang B, Kang HS, Koh C-S, Kim BS. Endothelin-1 contributes to the development of virus-induced demyelinating disease. *J Neuroinflammation*. 2020;17(1):307.
6. Ghanbari P, Khajehzadeh S, Sayyed A, Raeisi D, Salehi O. The effect of high intensity interval training with beetroot (*Beta vulgaris*) juice supplementation on serotonin and dopamine receptors expression, anxiety and depression in middle-aged diabetic rats. *Avicenna J Phytomedicine*. 2022;12(6).
7. Salehi O, Sheikholeslami-Vatani D, Hosseini SA. Positive effects of HIIT and vitamin E on cognitive function, angiogenesis and NRF2 antioxidant pathway in male rats with Alzheimer's disease. *Biogerontology*.

- 2025;26(4):129.
8. Einstein O, Katz A, Ben-Hur T. Physical exercise therapy for autoimmune neuroinflammation: Application of knowledge from animal models to patient care. *Autoimmun Rev.* 2022;21(4):103033.
 9. Sohrabi P, Parnow A, Jalili C. Treadmill aerobic training improve beam-walking test, up-regulate expression of main proteins of myelin and myelination in the hippocampus of cuprizone-fed mice. *Neurosci Lett.* 2023;792:136936.
 10. Goldberg Y, Segal S, Hamdi L, Nabat H, Fainstein N, Mediouni E, et al. High-intensity interval training attenuates development of autoimmune encephalomyelitis solely by systemic immunomodulation. *Sci Rep.* 2023;13(1):16513.
 11. Hamdi L, Nabat H, Goldberg Y, Fainstein N, Segal S, Mediouni E, et al. Exercise training alters autoimmune cell invasion into the brain in autoimmune encephalomyelitis. *Ann Clin Transl Neurol.* 2022;9(11):1792–806.
 12. Rezaee S, Kahrizi S, Nabavi SM, Hedayati M. VEGF and TNF- α Responses to Acute and Chronic Aerobic Exercise in the Patients with Multiple Sclerosis. *Asian J Sports Med.* 2020;11(3).
 13. Yaghoubi M, Kordi M, Gaeni A. The Effect of Forced and Voluntary Exercise before Induction of Experimental Autoimmune Encephalomyelitis on the Integrity of the Blood-Brain Barrier and Gene Expression of Some of Tight Junction Proteins. *J Appl Exerc Physiol.* 2020;16(32):87–101.
 14. Tajiri N, Yasuhara T, Shingo T, Kondo A, Yuan W, Kadota T, et al. Exercise exerts neuroprotective effects on Parkinson's disease model of rats. *Brain Res.* 2010;1310:200–7.
 15. Bernardes D, Oliveira ALR de. Regular exercise modifies histopathological outcomes of pharmacological treatment in experimental autoimmune encephalomyelitis. *Front Neurol.* 2018;9:950.
 16. D'Orléans-Juste P, Ndunge OBA, Desbiens L, Tanowitz HB, Desruisseaux MS. Endothelins in inflammatory neurological diseases. *Pharmacol Ther.* 2019;194:145–60.
 17. Yin X-X, Zheng X-R, Peng W, Wu M-L, Mao X-Y. Vascular endothelial growth factor (VEGF) as a vital target for brain inflammation during the COVID-19 outbreak. *ACS Chem Neurosci.* 2020;11(12):1704–5.
 18. Włodarczyk L, Cichoń N, Karbownik MS, Saso L, Saluk J, Miller E. Circulating serum VEGF, IGF-1 and MMP-9 and expression of their genes as potential prognostic markers of recovery in post-stroke rehabilitation—a prospective observational study. *Brain Sci.* 2023;13(6):846.
 19. Ali M, Falkenhain K, Njiru BN, Murtaza-Ali M, Ruiz-Urbe NE, Haft-Javaherian M, et al. VEGF signalling causes stalls in brain capillaries and reduces cerebral blood flow in Alzheimer's mice. *Brain.* 2022;145(4):1449–63.
 20. Yu Q, Xia Z, Liang EC, Tipoe GL. Chronic aerobic exercise improves insulin sensitivity and modulates Nrf2 and NF- κ B/I κ B α pathways in the skeletal muscle of rats fed with a high fat diet. *Mol Med Rep.* 2019;20(6):4963–72.
 21. Wenner MM, Welti LM, Dow CA, Greiner JJ, Stauffer BL, DeSouza CA. Aerobic exercise training reduces ET-1-mediated vasoconstriction and improves endothelium-dependent vasodilation in postmenopausal women. *Am J Physiol Circ Physiol.* 2023;324(6):H732–8.