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The preconditioning effect of different exercise training modes on middle cerebral artery occlusion induced-behavioral deficit in senescent rats

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ABSTRACT

Introduction: Brain abilities decrease after brain stroke in elderly. The neuroprotective effect of exercise training has been proved in clinical trials and animal experiment. Nevertheless, it is not still clear what kind of exercise has greater protective effect. The present study aimed at investigating pre-conditioning effect of endurance, resistance, and concurrent training on learning ability, anxiety, and spatial memory in aged rats following stroke strength with middle cerebral artery occlusion.

Method: We used 50 male Wistar rats (age = 24 months) that were assigned randomly in five groups; 1: sham group, 2: Control group 3: Endurance training 4: Resistance training, and 5: concurrent training. The exercise training groups received training for four weeks. Following training, middle cerebral artery occlusion was applied to induce cerebral ischemia. Using the elevated plus maze, shuttle box test, and Morris water maze, neurocognitive functions were tested in the sample rats.

Results: It was found that resistance training did not affect spatial memory in the acquisition phase, while concurrent training and endurance training enhanced spatial memory in the acquisition phase. On the contrary, spatial memory was improved by resistance training in the retention phase, while concurrent and endurance exercises did not affect spatial memory in the retention phase. Passive avoidance learning ability at acquisition phase was more in resistance group compared to the endurance and concurrent training in shuttle box test, but in retention phase was similar between training groups. Unlike endurance and concurrent training, resistance training reduced anxiety in senescent rats.

Conclusion: All three exercise types alleviated aversive learning and memory impairment induced by stroke in senescent rats. Notably, the resistance training showed a greater protective effect compared to the other two training methods.

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1. Introduction

Stroke as a severe medical problem has an incidence of 300–500 per 100,000, between the 45–84 ages [1]. Age is a critical determinant of cerebral ischemia and its recovery. Stroke is an important death factor and a cause of long-term disability in the world that is prevalent among the elderly [2]. As established by clinical medicine, people suffering from cerebral ischemia damage might develop a regional decline in neuronal cell loss, cerebral blood flow, cognitive and mood deficits [3–5]. There is an association between cognitive function impairment and elevated risk of functional outcomes and dementia [6]. Learning, memory, anxiety [4] as well as depression [7–9] have been identified as the most prevalent cognitive problems following stroke [4].

One of the main clinical predictors of functional outcome and mortality following a stroke is the severity of the stroke at admission [10]. If adjustable risk factors for severity of stroke at admission are understood, efficient strategies can be adopted to minimize post-stroke distress. An association has been found between physical activity (PA) and a reduction in stroke incidence in both males and females [11–14]. It has been proposed that PA can be a beneficial preconditioning driver [15] involved in the brain repair process following ischemia [16]. There are a large number of studies indicating the alleviatory effect of preconditioning through PA concerning cerebral-induced injuries [17,18]. As shown by review of research on animals, PA exerted a neuroprotective effect before stroke, which resulted in fewer neurological deficits and minimal stroke [19]. In another animal research, it was indicated that three weeks of bike exercise and a voluntary treadmill had useful effects in decreasing risk of stroke and presenting a prophylactic treatment approach to reduce brain injuries and increase blood flow during cerebral ischemia [20]. In a study by He et al. (2014), it was demonstrated that two-week treadmill pre-exercise decreased blood-brain barrier dysfunction during cerebral ischemia injuries and cerebral edema through the down-regulation of the aquaporin in rats [21]. An animal experiment was conducted by Feng et al. (2014) and it was shown that pre-ischemic treadmill exercise elevated superoxide dismutase action, decreased neurological deficits and infarct volume, and reduced the malondialdehyde concentration following ischemic stroke [22]. As suggested by Bovim et al. (2019), PA prior to stroke might not protect against post-stroke anxiety [23]. Nevertheless, as demonstrated by an animal study with middle cerebral artery occlusion (MACO), the pre-ischemic effect of voluntary wheel running beneficially affects levels of cognition, anxiety, and striatal dopamine in rats with cerebral ischemia [4]. In their study, Khabour et al. (2010) indicated the beneficial effect of voluntary exercise on long- and short-term spatial memory in rats and the elevation of hippocampal BDNF levels [24].

According to the above-mentioned studies, it is approved that pre-stroke PA is associated with post-stroke cognitive functions. Despite the popularity of PA as an appropriate approach to improve health outcomes, it is still not known which specific training approaches can exert a more protective effect on mood and cognitive functions. In the most PA training studies, aerobic training protocol was used for cognitive functions. It has been demonstrated that other kind of training protocol like resistance training may have positive effect on cognitive function, however the evidence is limit. For example, as shown by Cassilhas et al. (2012), eight weeks of resistance and aerobic training led to spatial memory function improvement in rats [25]. According to our knowledge there is no study that investigates the preconditioning effect of concurrent training (compounding resistance and aerobic training) protocol on cognitive function after brain stroke. Despite the establishment of a close relationship between ischemic brain injury and mood and cognitive function changes, it is not presently known whether different kinds of pre-ischemia exercise training play a protective function for neurocognition. Therefore for the first time, the present study aimed at investigating the impact of resistance, aerobic, and concurrent training on cognitive function in aged rats following stroke with middle cerebral artery occlusion.

2. Method and material

2.1. Subjects

Researchers at the animal laboratory prepared 50 male Wistar rats with 24-month age and weights between 350 and 400 g at Hamedan University of Medical Sciences. Standard conditions, including a humidity of 50 °C, a temperature of 22 °C, and a 12-h dark-light cycle, were applied for housing the animals. In addition, water and food were provided for the animals. Helsinki Convention was followed to work with animals and perform research methods. Animals were randomly grouped as following: 1: sham group (sham, n = 8), 2: Control group (Con + MCAO, n = 5) 3: Endurance training (En + MCAO, n = 5) 4: Resistance training (Res + MCAO, n = 5), and 5: concurrent training (Conc + MCAO, n = 5). It should be mentioned that, at first, 10 Wistar rats were selected in each group, but, 5 rats in each cerebral ischemia groups were died during and after middle cerebral arterial occlusion (MCAO), meanwhile, 2 rats were died in sham group during cerebral ischemia operation.

The Capital University Institutional Animal Ethics Committee approved all protocols and procedures.

2.2. Resistance training protocol

Strength training comprised climbing ladders. The railing height was 1 m with a slope of 85° and included 25 steps. The subjects were familiarized with ladder climbing one week prior to starting the training. The researchers conduct the exercises gradually for four weeks (five sessions per week). Subjects did the exercise in 4 sets of 6 repetitions in the first week with 3-min interval between each set, and the interval between each repetition was 15 s. The researcher attached lifting weight to the tails of the rats, holding 50% of their body weight in the first week, 60% of body weight in the second week, 70% of body weight in the third week, and 80% of body weight in the fourth week. The researcher measured the rat's weight weekly prior to starting the training, and the weight hanging from the subjects' tail specified [26].

2.3. Endurance training protocol

The animals got familiarized with the treadmill training in three sessions, lasting 10 min at a speed of 10 m/min. When the sessions were adjusted, rats practiced walking on a treadmill (5 s/week) for four weeks. In the first week, they performed the exercise for 15 min at a speed of 15 m/min. In the second week, they performed the exercise at a speed of 20 m/min for 20 min. In the two last weeks, the animals ran at a speed of 25 m/min for 25 min and at a speed of 30 m/min for 30 min [26].

2.4. Concurrent training protocol

Combined exercises comprised endurance and strength trainings concurrently. Half the aerobic exercises and half the strength exercises were performed in each session. In the first and third weeks, firstly, aerobic exercise was performed, and then strength exercise. In the second and fourth weeks, subjects performed aerobic exercise performed first, and then strength exercise. The first week comprised running at a pace of 15 m/min for 7.5 min, and after 5 min of complete rest, the subjects performed 4 sets of 3 repetitions with a weight of 50% body weight. In the second week, subjects performed 4 sets of 3 repetitions with a weight of 60% body weight, and after 5 min of complete rest, they worked at a speed of 20 m/min for 10 min. In the third week, the subjects ran 12.5 min at a speed of 25 m/min, and after 5 min of resting; they performed 4 sets of 3 repetitions with a weight of 70% of body weight and climbed the fence. Lastly, in the fourth week, they performed 4 sets of 3 repetitions of 80. They performed 100 bodyweights for 5 min, followed by running at 30 m/min for 15 min [26].

The sham group and control group did not have any activities during this period.

2.5. Middle cerebral artery occlusion

Subjects endured cerebral ischemia surgery 72 h after the last exercise session. The researchers did temporary occlusion of the brain's middle cerebral artery. Firstly, they applied intraperitoneal injection of - xylazine (15 mg/kg) - ketamine (60 mg/kg) to anesthetize the rats. Then, after shaving the midsection of the neck, they used a 16-gauge razor blade to make a 2 cm by 2 mm wound to the right of the neck midline. Afterward, they gently pushed aside the muscles and the membrane torn. Then, the researcher gently separated the common carotid artery from the vagus nerves without making damage to it. The distal part of the external common carotid artery was closed completely with 0.6 silk threads. In addition, a blood vessel clamp was used to temporary closing the proximal section of the common carotid artery. Similarly, using a clamp, the distal portion of the internal carotid artery was blocked temporarily. A very thin wound was created in the blood vessel wall of the external common carotid artery adjacent to the junction of the common carotid artery. It was followed by inserting a microfilament (a length of 5 cm and a diameter of 0.2 mm) into the external common carotid artery (after removal of the clamp). It was conducted to the brain. After entering approximately 20 mm of filament into the artery, the filament showed relative resistance, which indicated middle cerebral artery obstruction. The filament was kept in this position for 30 min for inducing ischemia. It was slowly taken out after 30 min and brain reperfusion was restored. Lastly, after suturing the neck skin, the subjects were individually boxed for recovery until regaining full consciousness [27].

In the sham group the distal section of the external common carotid artery was obstructed and the cervical area was opened. However, there was no penetration of the filament into the internal common carotid artery.

2.5.1. Behavioral tests

We conducted behavioral tests one day after the rats regained their consciousness for ensuring cerebral ischemia. For this purpose, rats were assessed using movement tests (rotating their girth on a flat surface), hanging by the tail, reflex (corneal reflex, startle reflex, and pinna reflex), and balance test (beam balance test). Based on the complication severity, a score between 0 and 18 was given. A score of 0 implies no cerebral ischemia, 1 to 6 as mild ischemia, 7 to 12 as moderate ischemia, and 13 to 18 as severe ischemia. One day after confirming a stroke, rats performed Abbey Morris maze test.

2.6. Morris water maze

Using the Morris water maze, learning ability and spatial memory were measured. It is a circular pool with a height of 60 cm and a diameter of 180 cm, in black color, which is filled with water to 25 cm depth. The water temperature was $22 \pm 1^{\circ}$ Celsius. Using dim lights, the room was lit and it was soundproof. We issued some visual cues. In each pool quadrant, 4 starting lines were presented, like West (W), South (S), North (N), and East (E). In quadrant N is an invisible Plexiglas platform with a diameter of 10 cm that is located centrally 1 cm under the water. The researcher trained subjects for 4 days at approximately the same time. They performed two blocks of 4 attempts each day (90s). Between every two tests, the subjects rested on the platform for 30 s. Furthermore, a 5-min gap was given between two consecutive sessions. The researchers recorded the time for reaching the hidden platform (escape latency) using an embedded video camera (New York, USA). They connected camera directly to the PC above the pool. The spatial acquisition phase was performed within 4 days. The retention phase was conducted on the fifth day. Subjects conducted a probe attempt in the retention phase, in which we excluded the hidden platform from the pool. In this case, the animals were allowed swimming for 60 s in order to record the swimming speed and the time ratio spent in the target quadrant [28].

2.7. Shuttle box

One day following the Water Morris maze test, the researchers evaluated cognitive function using the shuttle box test and a stepwise passive avoidance task. It an automated shuttle box with dark and lighted sections of the same size, which was separated by a stainless bar screen as a base and a sliding door. After the adaptation phase, the researchers performed a single experiment. To this end, they put the animals in the lighted section for 2 min and the door to the dark section closed. After a period of adaption for 2 min, the dark section door was opened automatically for the learning experiment. After rats entering the dark section, the lighted section door was closed automatically. Then, a single inevitable, coded foot shock (0.2 mA, 2 s) was carried via the grid base. The process was repeated until the latency entering the dark section was above 300 s. 24 h after the learning test, we measured the memory test. The researcher put rats in the lighted section, with opening the door after 2 min. They documented the latency for entering the darkroom with a maximum of 600 s. After performing the retention test, there was no foot shock. Rats that not entering the darkroom in the retention test were allocated a latency time of 600 s [28].

2.8. Elevated plus maze

The elevated plus maze test was carried out one day after the shuttle box. Using this test, the anxiety-like behavior was analyzed in rats with cerebral ischemia (22/35). The EPM contained black polypropylene with two opposite closed arms (25 × 8 × 20 cm), two opposite open arms (25 × 8 cm), and a central platform (8 × 8 × 8 cm) as a cross. The maze was elevated 50 cm above the ground. The subjects individually seated in the center and their heads directed toward one of the closed arms. The number and time of inputs in the closed and open arms were recorded during a test period of 300 s. Entry with closed or open arms was described as entry of four paws into one arm. Following the behavioral test, the researcher cleaned the maze with 75% ethyl alcohol solution, letting to be dried between tests [28]. Experimental time line was shown in figure-1.

2.9. Statistical analysis

GraphPad Prism 9 software (GraphPad Software, San Diego, CA, USA)utilized for statistical analysis and graphs generate. Using a two-way repeated measures test, the difference between groups in escape latency in the MWM test assessed, and one-way ANOVA followed by Tukey's post-hoc test employed for other variables. Data given as Mean ± SEM. A significance level below 0.05 considered.

3. Results

Modified nerve severity score (mNSS) in one day after MCAO induction was different between experimental groups ($F = 37.5$, $p = 0.0001$). As shown in Fig. 2, mNSS in control group was more compared to other study groups ($p < 0.05$). Furthermore, mNSS significantly decreased in response to different exercise training, but was not differ between training groups ($p > 0.05$).

3.1. Morris water maze

Mean of scape latency. The results of ANOVA test showed that there is a significant difference between groups in 4 days of study ($F = 5.66$, $p = 0.012$). Comparison of the changes within the groups showed that in all five groups the time to find the platform significantly reduced on the third and fourth days compared to the first day ($p < 0.05$). Also, the comparison between the groups showed that the time to find the platform was significantly longer in the control group than in the sham group on the second, third and fourth day ($p < 0.01$). The resistance training group was not significantly different from the control group ($p = 0.7$), but the time to find the platform was significantly shorter in the endurance training group ($p = 0.018$) and concurrent training group ($p = 0.037$) than in the control group, but there was no significant difference with the sham group ($p = 0.4$). It should be noted that there was no significant difference between the endurance group and concurrent training group ($P = 0.9$) (Fig. 3a). In this regard, the results showed that there was a significant difference in the mean of total escape latency ($F = 8.3$, $p = 0.0003$). The mean of total escape latency in the control group was higher than the sham group ($p = 0.0008$). The resistance group was not significantly different from the control group ($p = 0.1$). Also, concurrent group ($p = 0.002$) and endurance group ($p = 0.0006$) were significantly less than the control group (Fig. 3b).

In addition, the results showed that the percentage of time spent in the target quarter on the day of the probe was significantly

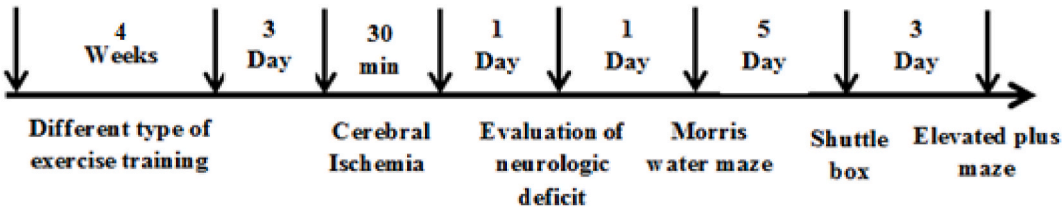

Fig. 1. Experimental time line.

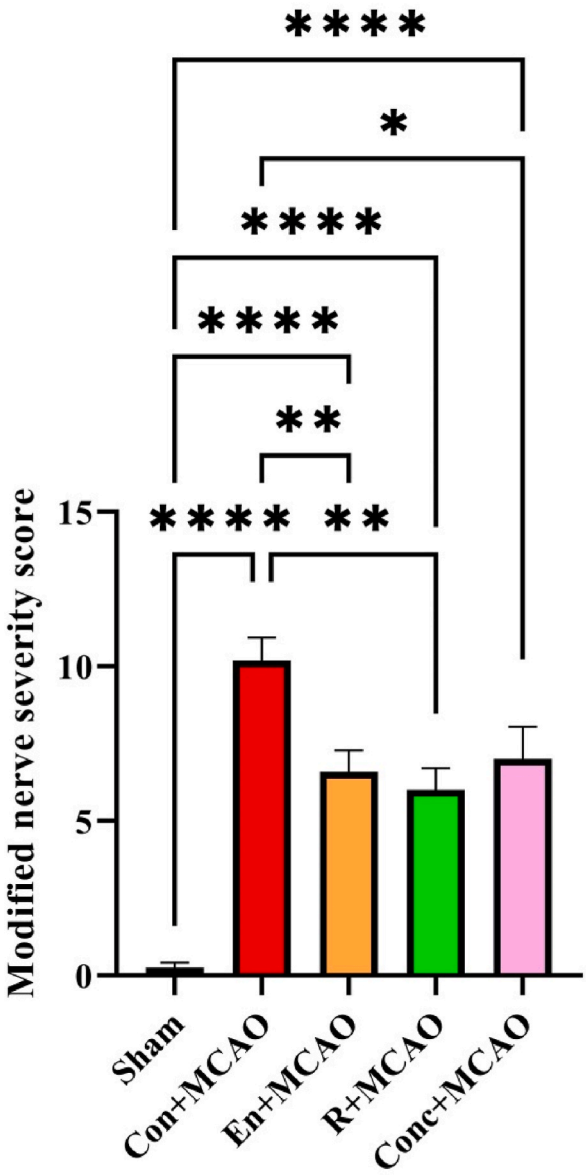

Fig. 2. Modified nerve severity score at 1 day after MCAO induction was different between experimental groups. Data are expressed as Mean ± SEM. *p < 0.05, **p < 0.01, ****p < 0.0001.

different between study groups (F = 13.5, p < 0.0001). In the control group the percent of time spent in the target quadrant was significantly lower than the sham group (p < 0.0001). Time spent in target quadrant at probe day in endurance (p = 0.02), resistance (p < 0.0001) and concurrent (p = 0.03) groups was more than control group. The results also showed that resistance training group performed better than other groups, which was higher than aerobic (p = 0.055) and concurrent (p = 0.04) groups. There was no significant difference between concurrent and endurance training groups (Fig. 3c).

3.2. Shuttle box

As shown in Fig. 4a, there is no significant difference between groups in number of trials (F = 0.27, P = 0.8). Furthermore, results showed that Step-Through Latency in acquisition phase (STLa) was significantly different between experimental groups (F = 7.45, p = 0.0005). STLa was higher in the control group than sham group (p = 0.0004). The results also showed that endurance training (p = 0.36) resistance training (p = 0.006) and concurrent training (p = 0.13) reduced STLa compare to the control group. There was no significant difference between training groups (Fig. 4b). Furthermore, STLr was different between experimental groups (F = 22.6, p < 0.0001). STLr in control group was significantly lower than other groups (p < 0.0001). STLr was not different between training groups.

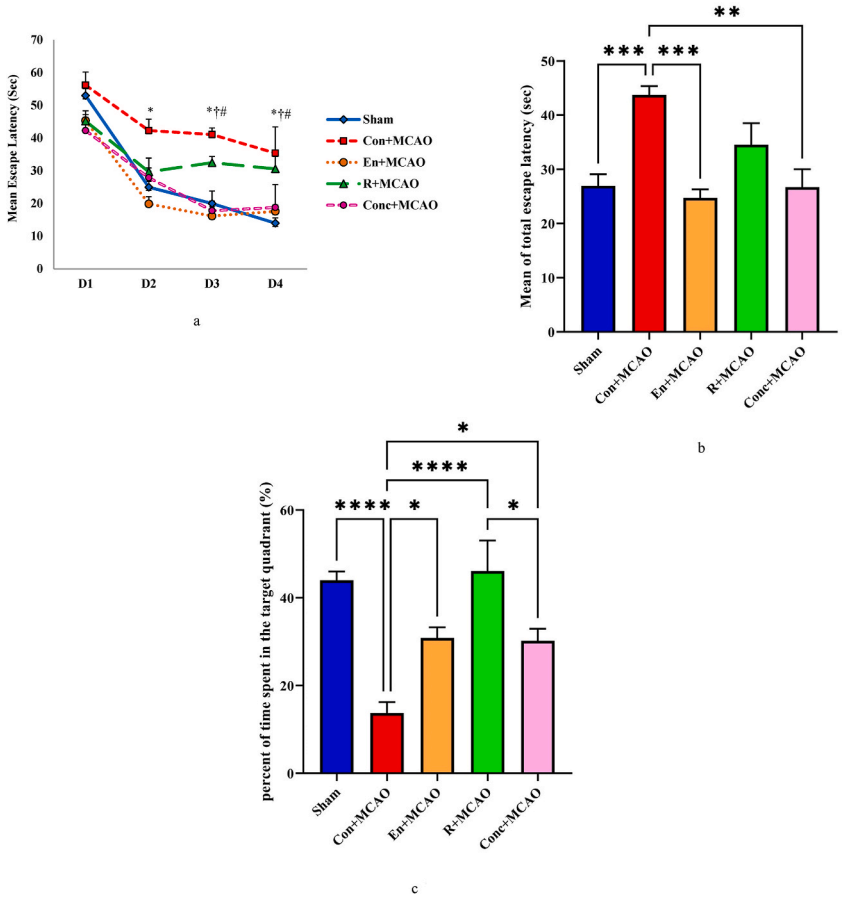

Fig. 3. A Comparison of the different exercise training modes to find the hidden platform at four days of acquisition phase in the Morris Water Maze test. Data are expressed as Mean ± SEM. * = significant difference vs. control group.† = significant difference vs. resistance group. # = significant difference vs. first day (D1).

B. Mean of total escape latency at four days of acquisition phase in the Morris Water Maze test was different between experimental groups. Data are expressed as Mean ± SEM. **p < 0.01, ***p < 0.001.

C. Percentage of time spent in the target quadrant in the probe day was different between experimental groups in the Morris Water Maze test. Data are expressed as Mean ± SEM. *p < 0.05, ****p < 0.0001.

It should to be noted that STLr in training groups was not different compared to the sham group (Fig. 4c).

Also, the results showed that there was a significant difference between research groups ($F = 42.4$, $p < 0.0001$) in time spent in the dark compartment (TDC) in the passive avoidance, so that the TDC in control group was significantly higher than sham group ($p < 0.0001$). All exercise groups showed reduced TDC compared to the control group ($p < 0.0001$), but there was significant difference between the training groups and the sham group. It should to be mentioned that TDC was not differ between training groups (Fig. 4d).

3.3. Elevated plus maze

As shown in Fig. 5, percent of time spent in close arm in elevated plus maze test was significantly different between groups ($F = 9.41$, $p = 0.0001$). Anxiety was not differ between sham and control groups ($p = 0.9$). This result indicated that anxiety was not affected by cerebral ischemia in short time. On the other hands, unlike endurance and concurrent training, resistance training significantly decreased anxiety in senescent rats with cerebral ischemia ($p = 0.0018$).

4. Discussion

Cerebral ischemia is generally deadly, with a limited treatment prognosis. Thus, using early prevention, the stroke onset risk can be reduced [29]. It has been proposed that physical exercise has neuroprotective effects in clinical trials and animal experiments [30]. Nevertheless, it is still unclear that which kinds of exercises could exert a greater protective effect on cognitive functions after cerebral ischemia in aging. Presently, some types of exercise training protocols exist, including resistance, endurance, and concurrent training protocol that are well-known in the literature related to exercise training [26,28]. Hence, the present study aimed at investigating the

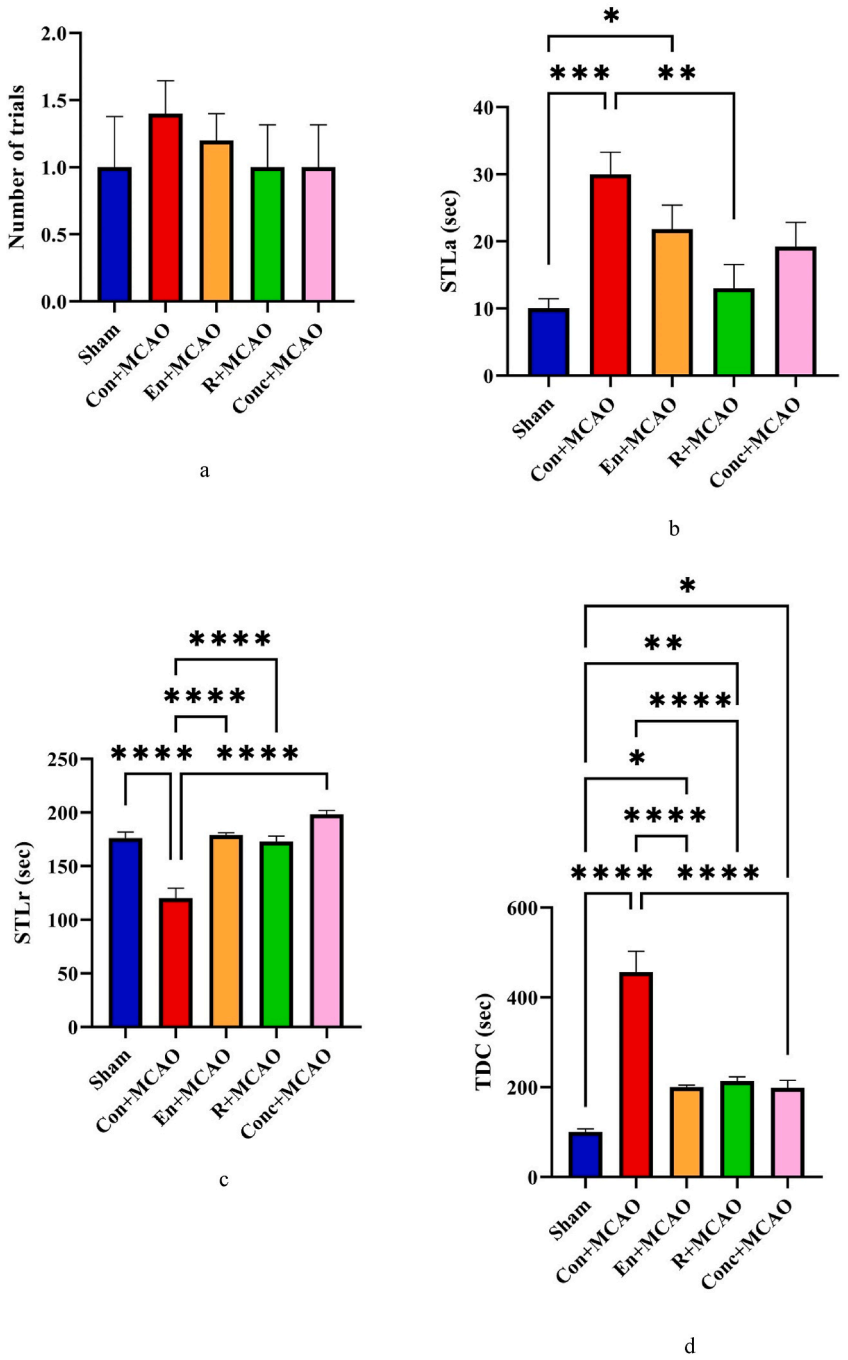

Fig. 4. A Number of trials to reach learning in passive avoidance learning in shuttle box test was not different between experimental groups. Data are expressed as Mean ± SEM.

B. Passive avoidance learning in acquisition phase (STLa) was significantly different between experimental groups. Data are expressed as Mean ± SEM. *$p < 0.05$, **$p < 0.01$, ***$p < 0.001$.

C. The entrance latency to the dark section in retention phase (Step-Through Latency, STLr) in passive avoidance learning was different between groups. Data are expressed as Mean ± SEM. ****$p < 0.0001$.

D. Time spent in the dark compartment (TDC) in the passive avoidance memory in retention phase was shorter in training groups compared to the control group. Data are expressed as Mean ± SEM. *$p < 0.05$, **$p < 0.01$, ****$p < 0.0001$.

effect of resistance, aerobic, and concurrent exercise on cognitive function (learning, anxiety, stress) in post-stroke aged rats with middle cerebral artery occlusion.

The results of this study showed that cognitive deficit significantly promote after middle cerebral artery occlusion in senescent rats.

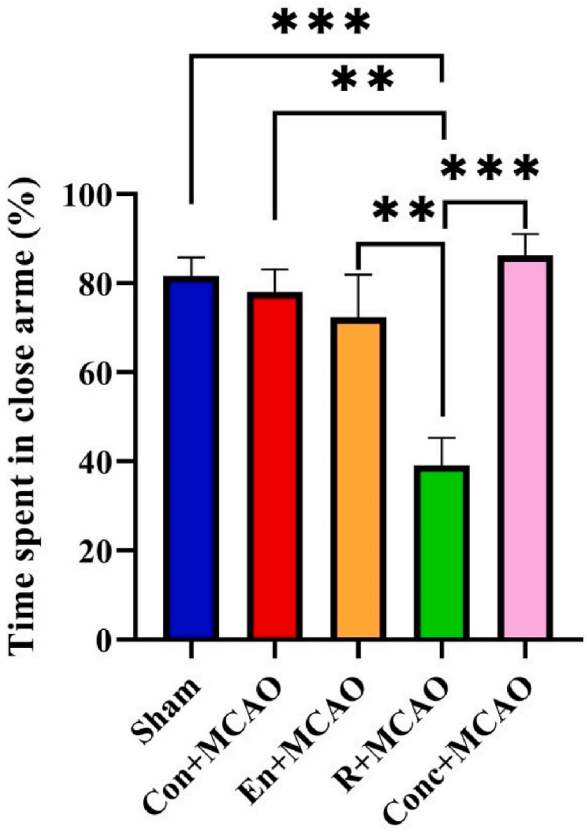

Fig. 5. Unlike endurance and concurrent training, resistance training significantly decreased anxiety in senescent rats. Data are expressed as Mean ± SEM. **p < 0.01, ***p < 0.001.

This finding is in line with previous studies that showed cerebral stroke causes behavioral impairment in senescent rats [31,32]. Briefly, the results of this study showed that compared to the other two methods, resistance training has less effect on improving spatial memory in the acquisition phase, and it was while resistance training in retention phase was more affective in spatial memory amelioration compare to the aerobic and concurrent training. It should to be noted that passive avoidance learning amelioration in acquisition phase in resistance training groups was more compare to the aerobic and concurrent groups, but in retention phase passive avoidance memory was similar between different training modes. Also, Stroke-induced anxiety was significantly reduced in response to resistance training. As demonstrated by our findings, all exercise types caused the mitigation of memory impairment and aversive learning induced by stroke. Regular exercise can improve neurogenesis, synaptic plasticity, and angiogenesis [33–35]. With exercise, blood flow to the brain increases, facilitating the metabolic waste elimination [35]. Specially, our results indicated the lack of effect for resistance training on spatial memory in the acquisition phase, while concurrent training and endurance training enhanced spatial memory in this phase. According to our knowledge, there has no study that compares protective effect of different types of exercise on cerebral ischemia induced behavioral disorders, but, our laboratory previously reported that varied types of exercise have diverse impacts on different learning and memory capabilities in morphine dependent rats [26]. Zarrinkalam et al. reported that different types of exercise have similar effects on spatial memory, but have different effects on passive memory after morphine withdrawal in rats [26]. They demonstrated that physical activity, regardless of type, blocks morphine-induced behavioral deficit.

Endurance exercise improved cognitive functions in rodents with cerebral ischemia by the stimulation of synaptic plasticity, angiogenesis, and neurogenesis through upregulating neurotrophin levels [36–39]. It has been demonstrated that preconditioning endurance exercise-induced neuroprotection influences some systems, such as cerebral vasculature remodeling, neurogenesis and angiogenesis stimulation, modulation of excitatory signals, preservation of blood-brain barrier integrity, and decrease of inflammatory markers [40]. Neuroprotection induced by stress could also be helpful through upregulated HSP 70, HSP 20, pERK 1/2, and other molecules, which concurrently disrupt cell death and promote survival signals. It is thought that glial cells, e.g., astrocytes, possibly have a role in aggravating secondary brain injury after neurotrauma [41].

On the contrary, our findings indicated that strength training caused spatial memory improvement in the retention phase, while concurrent and endurance training did not affect spatial memory. In addition, Cassilhas et el. (2012) found a positive impact of resistance training on spatial memory. It was indicated that resistance exercise reduced hippocampal and peripheral IGF-1 with concurrent activation of AKT and IGF_1 receptor in the hippocampus, which culminated ab elevation in the expression of synaptophysin and synapsin-1 [25]. As shown by Cassilha et al. (2012), aerobic exercise positively affected the spatial memory of rats in

varying molecular pathways, which is inconsistent with the findings of the present study concerning endurance training. Besides, our findings in the retention phase showed inconsistency with the findings Fan et al. (2021) and by Khabor et al. (2010). Gomez et al. (2015) found a positive impact of voluntary aerobic exercise on spatial memory since it elevated the BDNF and striatal dopamine levels in the hippocampus. Brain-derived neurotrophic factor (BDNF) is a small dimeric protein, functioning via tyrosine kinase B (TrkB) as its receptor. BDNF causes the modulation of neuronal cell growth and survival, and it is involved in memory and learning processes. Thus, BDNF dysfunction is associated with cognitive deficits [42]. BDNF improves long-term potentiation and hippocampal-dependent memory through TrkB [43]. The concentration of BDNF is high in the hippocampus, and the expression of BDNF is elevated selectively following activity-dependent memory and learning tasks [44]. It has been indicated that the activation of TrkB has an inhibitory effect on apoptosis following subarachnoid hemorrhage [45]. Khabour et al. (2010) and Cassilhas et al. (2012) conducted aerobic training for 6 and 8 weeks, while endurance training performed for 4 weeks in our work. It seems that molecular changes in the brain, which influence learning and memory, need longer training time, at least 6 weeks.

Additionally, the present work also aimed at investigating the mood function changes following cerebral ischemia with preconditioned resistance, endurance, and concurrent exercise. Although anxiety significantly reduced after resistance training, no difference occurred in behaviors related to anxiety between the case groups. It is inconsistent with the findings by Gokdemir et al. (2019) and Fan et al. (2021). Some discrepancies exist between the findings of the present work and their results. As an example, they did voluntary endurance exercise while it was not as the same in our study. Besides, their studies applied a longer exercise protocol time compared to our work. Khabour et al. (2010) indicated an increase in hippocampal BDNF following six weeks of voluntary aerobic training, and growing evidence indicates the crucial role of the declined brain BDNF levels in the pathogenesis of mood disorders and in the mechanism of action of treatments, including antidepressants and mood stabilizers [46,47]. The time period considered in our work for the training protocol seems to be inadequate for increasing the BDNF levels so that protective effect is generated against mood disorders following stroke in rats. Besides, it has been indicated that changes in the serotonergic system have a potential contribution to depression and anxiety. Brain regions, such as the amygdala, hippocampus, and frontal cortex, have declined serotonin levels, while serotonin receptor 1 A (5-HT-1) is elevated in depression and anxiety [48]. According to previous works, noticeable fluctuations were found in brain dopamine secretion in rats with global ischemia [49]. Additionally, dopamine is a neurotransmitter contributed to behavioral responses with a critical role in anxiety [50]. Also, there are a large number of studies showing a considerable change in emotional and cognitive function induced by dopamine degeneration through treating striatal dopamine pathways [51,52]. There is a close relationship between the striatum function and learning ability, motor control, memory ability, action selection, emotional adaptation, and stimulus-response learning [53]. Moreover, the striatum is a significant area vulnerable to ischemic insult [54]. As shown by previous research works, cerebral ischemia can result in dopamine release in the striatum [55]. Besides, a study showed the occurrence of the elevated dopamine release in the striatum during bilateral carotid artery clamping [56]. Brain injury is reduced by exercise preconditioning through the reduction of cerebral permeability and improvement of brain integrity after stroke [57]. As confirmed by some studies, the antioxidant ability can be increased by exercise preconditioning, leading to a significant promotion of emotional and cognitive function [22,58].

The findings of this study should be considered in light of its limitations. Firstly, the mechanism (molecular or structural) underlying the neuroprotective effects of aerobic, resistance, and concurrent training on cognitive function in post-stroke aged rats with middle cerebral artery occlusion of should be considered in future studies. It has been indicated that pyramidal neurons of hippocampus CA1 region among brain neurons show sensitivity to ischemia states [59]. The role of these neurons is critical in memory and learning functions and affect by training. So future studies can consider this region of brain. Secondly, the short-term training period was used in this study. We propose to future studies to investigate the long-term effect of these training protocols.

5. Conclusion

In general, the results of this study showed that different exercise training modes have varying affects on middle cerebral arterial occlusion–induced behavioral disorders in senescent rats. All three training methods are an effective strategy in the treatment of cerebral ischemia-induced behavioral deficit, but resistance training seems to be more effective.

Author contribution statement

Ebrahim Zarrinkalam: Performed the experiments; Wrote the paper.
Seyedeh Manizheh Arabi: Analyzed and interpreted the data; Wrote the paper.
Alireza Komaki: Conceived and designed the experiments; Wrote the paper.
Kamal Ranjbar: Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.

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Data availability statement

Data will be made available on request.

Ethics approval and consent to participate

All experiments were conducted based on the National Institutes of Health and the Guide for the Care and Use of Laboratory Animals (NIH Publication 80–23, 1996). The Ethical Committee of Hamadan University of Medical Sciences confirmed the research protocol. Full efforts were made to reduce animals' use and advance their welfare.

Consent for publication

All authors are agreed to publish this manuscript.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Not applicable.

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