Research article

Resistance training boosts lactate transporters and synaptic proteins in insulin-resistance mice

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A B S T R A C T

Background: This investigation delineates the influence of resistance training on the expression of synaptic plasticity-related proteins in the hippocampi of insulin-resistant mice and explores the underlying molecular mechanisms.
Methods: Six-week-old male C57BL/6 J mice were stratified into a control group and a high-fat diet group to induce insulin resistance over a 12-week period. Subsequently, the mice were further divided into sedentary and resistance training cohorts, with the latter engaging in a 12-week ladder-climbing regimen. Post-intervention, blood, and hippocampal specimens were harvested for analytical evaluation.
Results: In the insulin-resistant mice, elevated blood lactate levels were observed alongside diminished expression of synaptic plasticity-related proteins, monocarboxylate transporters (MCTs), and reduced phosphorylation of protein kinase B (Akt) and mechanistic target of rapamycin (mTOR). In contrast, the expression of eukaryotic translation initiation factor 4 E-binding protein 2 was significantly augmented. Resistance training mitigated insulin resistance, decreased blood lactate levels, and enhanced the expression and phosphorylation of mTOR, regulatory-associated protein of mTOR, MCTs, and synaptic plasticity-related proteins.
Conclusions: Resistance training mitigates insulin resistance and improves hippocampal synaptic plasticity by normalizing blood lactate levels and enhancing mTOR, MCTs, and synaptic plasticity-related proteins. It may also activate mTORC1 via the PI3K/Akt pathway, promote lactate utilization, and enhance synaptic plasticity proteins, potentially alleviating peripheral insulin resistance. Further research is needed to confirm these mechanisms.

1. Introduction

Numerous investigations have established a robust association between insulin resistance and cognitive deficits [1–4]. Peripheral insulin resistance is known to suppress the phosphatidylinositol 3-kinase/protein kinase B (PI3K/Akt) signaling pathway within the central hippocampus [5], adversely affecting the mechanistic target of rapamycin complex 1 (mTORC1). This suppression leads to diminished activity of eukaryotic translation initiation factor 4 E-binding protein (4EBP) and ribosomal protein S6 kinase (P70S6 K), culminating in reduced expression of synaptic plasticity-related proteins such as *N*-methyl-D-aspartate receptor (NMDAR) and synapsin (SYN). These molecular alterations negatively impact learning and memory functions [6,7]. Furthermore, insulin resistance or type 2

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diabetes diminishes the organism's oxidative capacity, elevating blood lactate levels and perpetuating insulin resistance in a self-reinforcing cycle [8]. In scenarios of compromised oxidative capacity, brain lactate serves as a vital energy substrate to maintain normal brain functionality [9,10], a process facilitated by monocarboxylate transporters (MCTs). MCT1 and MCT4 are involved in the shuttling of peripheral blood lactate into the brain or its exportation from astrocytes, whereas MCT2 on neuronal membranes mediates the transport of lactate from the interstitium into neurons for oxidative metabolism [11]. Any disruption in this lactate transport mechanism can lead to decreased brain adenosine triphosphate levels, further inhibiting mTORC1 activity and ultimately impeding protein synthesis [12].

Exercise has been shown to ameliorate insulin resistance [13,14], bolster hippocampal neuronal synaptic plasticity, and enhance learning and memory functions, thus mitigating and preventing cognitive deficits [15,16]. Furthermore, physical activity elevates the expression of synaptic plasticity-related proteins within the hippocampus of individuals with insulin resistance [17]. While resistance training has been demonstrated to activate the PI3K/Akt signaling pathway in the hippocampus of normal Wistar rats [18], research on the effects of resistance exercise on the hippocampal PI3K/Akt signaling pathway and lactate transport mechanisms in models of insulin resistance remains scarce.

The objective of this study is to examine alterations in the PI3K/Akt signaling pathway and synaptic plasticity-related proteins in the hippocampus of insulin-resistant mice. Additionally, this research aims to determine whether a 12-week regimen of resistance exercise can ameliorate the PI3K/Akt signaling and lactate transport dynamics in the hippocampus of IR mice, thereby augmenting synaptic plasticity. We hypothesize that resistance training will rectify insulin signaling disturbances and enhance synaptic plasticity in the hippocampus of these mice.

To address these aims, we developed a mouse model of insulin resistance through a high-fat diet and implemented resistance training as a therapeutic intervention. Changes in synaptic plasticity were assessed by measuring the expression of synaptic plasticity-related proteins. Furthermore, blood lactate levels, the activity of the PI3K/Akt pathway, and the expression of hippocampal MCT proteins were evaluated to elucidate the underlying molecular mechanisms.

2. Materials and methods

2.1. Experimental animals

The study utilized 6-week-old male C57BL/6 J mice obtained from the Nanjing Model Animal Research Center. The mice were housed in a controlled environment with a 12-h light/dark cycle at a room temperature of (22 ± 2) °C and a relative humidity of 40~70 %. Adequate food and water were provided to the mice ad libitum throughout the experimental period. Ethical approval was obtained from the relevant ethics committee at the institution. All necessary measures were taken to minimize the number of animals used and to reduce potential suffering.

2.2. Establishment of animal model and grouping

After one week of acclimation to adjustable feeding, the mice were randomly divided into two groups: the normal control group (C, n = 12) and the high-fat diet group (HFD, n = 26). They were subjected to their respective diets for 12 weeks. Following the dietary intervention, both the control and high-fat diet groups underwent an overnight fast, and blood samples were collected from the tail vein the next day. The glucose tolerance test (GTT) and insulin tolerance test (ITT) results were utilized to confirm the successful establishment of the insulin resistance model (based on the significant difference in the area under the blood glucose level-time curves between the HFD and C groups [19]). Subsequently, the mice with insulin resistance were randomly assigned to two groups: the insulin resistance sedentary group (IR, n = 10) and the resistance training group (RT, n = 10).

2.3. Exercise protocols and diets

Mice in group C and group IR did not exercise, and mice in group RT performed a 12-week increasing load ladder-climbing training after being adapted to the ladder for a week. The resistance training was carried out with a weight-climbing ladder with a ground inclination of 85°, a step interval of 2 cm, and a height of 1 m, 5 times/group (1 min interval), 3 groups/day (group interval 2 min), 3 days/week. The exercise protocols were based on the research of Leite et al. [20]. The training load is arranged as follows: the first week, the load was 30 % of the mouse's own body weight, and gradually increased to 110 % of the mouse's own body weight in the 12th week. Exhaustion was determined when the animal could not progress up the ladder after three successive stimuli to the tail, and then stopped training. During the training period, every group of mice maintained their original way of feed. (Table 1).

Table 1
Composition of normal diet and high fat diet.

	Normal diet	High fat diet
	kcal%	kcal%
Protein	24.6	20
Fat	12.5	60
Carbohydrates	62.9	20

2.4. Insulin sensitivity test

For the GTT, after an overnight fast, a 50 % glucose solution was administered intraperitoneally at a dosage of 2 g/kg body weight. Blood glucose concentrations were measured from tail vein samples at intervals of 0, 15, 30, 60, 90, and 120 min post-injection. For the ITT, following an overnight fast, insulin was injected intraperitoneally at a dose of 1 U/kg body weight, and blood glucose levels were similarly assessed at the same time points. The temporal progression was plotted on the x-axis against blood glucose concentration on the y-axis to generate segmented line graphs for both GTT and ITT. The area under the curve (AUC) for each test was calculated. A higher AUC indicates reduced insulin sensitivity. Both GTT and ITT were conducted twice, following 12 weeks on a high-fat diet and after 12 weeks of resistance training intervention.

2.5. Biochemical examination of the blood

Blood was taken from the heart. After 48 h of the end of the 12-week resistance training intervention, all mice were anesthetized with 10 % chloral hydrate (the anesthetic dose was 4 ml/kg body weight) and sacrificed. The blood was quickly taken from the heart to prepare the plasma samples. Plasma samples were sent to the Nanjing Institute of Bioengineering to test the plasma lactate levels of each group of mice.

2.6. Hippocampal proteins extraction

After 12 weeks of resistance training intervention and 48 h after the last training session, all the mice were anesthetized with isoflurane and sacrificed, and the brains were quickly taken. The hippocampi were separated from the brain on ice, stored in an EP tube, and quickly stored in a refrigerator at $-80\ ^{\circ}$C.

The hippocampus of each mouse was washed in a 2 ml tube with pre-cooling PBS. The hippocampus was centrifuged at 600 rpm for 30 s for one time, and the supernatant was removed. Then, the hippocampus of each mouse was fully cut into pieces in a 2 ml tube with the mixture (500 µl RIPA, 5 µl PMSF, and 55.5 µl phosphatase inhibitor), and the protein sample was circulating extracted with ultrasonic wave (20 s × 2 times). Afterward, the mixture was centrifuged (12000 rpm × 20 min, 4 $^{\circ}$C) to collect the supernatant, which was hippocampal proteins. The protein concentration was determined by the BCA method (BCA Protein Assay Kit, WB0125, Biotech Well, Shanghai, China). According to the concentration determination result, a certain amount of protein loading buffer (5 ×) and PBS were added to adjust the concentration of the sample to 4 µg/µl. The protein loading buffer was simultaneously diluted to 1 × , and the diluted sample was boiled at 95 $^{\circ}$C for 5 min to denature the protein. The prepared samples were stored at $-80\ ^{\circ}$C.

2.7. Western blot

The protein samples that contained an equal amount of protein (32 µg) were electrophoresed on 10 % sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE) gels and transferred to polyvinylidene fluoride (PVDF) membranes. The membranes were blocked with 5 % non-fat milk powder or bovine serum albumin (BSA) in TBST buffer and incubated overnight at 4 $^{\circ}$C with different primary antibodies. PI3K (1:1000, #4292, Cell Signaling Technology, MA, USA), Phospho-PI3K(1:1000, #4228 S, Cell Signaling Technology, MA, USA), Akt (1:1000, #9272, Cell Signaling Technology, MA, USA), Phospho-Akt (1:1000, #4060 S Cell Signaling Technology, MA, USA), mTOR (1:1000, #2972 S, Cell Signaling Technology, MA, USA), phosphor-mTOR (1:1000, #2971 S, Cell Signaling Technology, MA, USA), 4EBP2 (1:500, #2845 S, Cell Signaling Technology, MA, USA), Phospho-4EBP2 (1:200, #2855 S, Cell Signaling Technology, MA, USA), Raptor (1:1000, #2280 S, Cell Signaling Technology, MA, USA), P70S6 K (1:1000, #34475 S, Cell Signaling Technology, MA, USA), Phospho-P70S6 K (1:200, #9234 S, Cell Signaling Technology, MA, USA), postsynaptic density protein 95 (PSD95) (1:1000, #3450 S, Cell Signaling Technology, MA, USA), NMDAR(1:1000, #5704 S, Cell Signaling Technology, MA, USA), MCT1(1:1000, ab93048, Abcam, Cambridge, UK), glucose transporters (GLUT1) (1:100000, ab115730, Abcam, Cambridge, UK), GLUT3(1:200, ab315168, Abcam, Cambridge, UK), SYN(1:1000, 4329 S, Cell Signaling Technology, MA, USA), MCT2(1:500, sc-166925, Santa Cruz Biotechnology CA, USA) and MCT4 (1:500, sc-376140, Santa Cruz Biotechnology CA, USA) were used to detect the Hippocampal proteins, and β-Actin (1:10000, 66009-1-Ig, Proteintech, Chicago, USA) and β-Tubulin (1:5000, 10068-1-AP, Proteintech, Chicago, USA) were used as the loading control. After rinsing with TBST, the membranes were incubated with secondary antibody peroxidase-conjugated goat anti-rabbit and goat anti-mouse IgG (H + L) (1:5000, SA00001-1/2, Proteintech, Chicago, USA) for 1 h at RT. To visualize the immunoreactive protein bands, an advanced reagent (Millipore, USA) and an automatic chemiluminescence apparatus were used according to the manufacturer's instructions. The density of bands was determined by Image J software.

2.8. Statistical analysis

The results of each protein index were expressed as mean ± standard deviation ($\bar{X} \pm SD$). All data were statistically analyzed using SPSS 20.0 data processing software. The independent sample T test was used for comparison between group C and group HFD. One-way analysis of variance (ANOVA) was used in comparison between group C and group IR, group IR and group RT. The difference was significant at $P < 0.05$. *$P < 0.05$, **$P < 0.01$ vs. C; #$P < 0.05$, ##$P < 0.01$ vs. IR.

3. Results

3.1. Effects of high-fat diet and resistance training on insulin sensitivity in mice

After 12 weeks of a high-fat diet, GTT and ITT were performed. The AUC of GTT and ITT in the HFD was significantly higher than that in the normal control group (C) ($P < 0.05$, Fig. 1A–D), indicating the successful establishment of the insulin resistance model. Following 12 weeks of resistance training intervention, the AUC value of the IR was significantly higher than that of group C ($P < 0.01$).
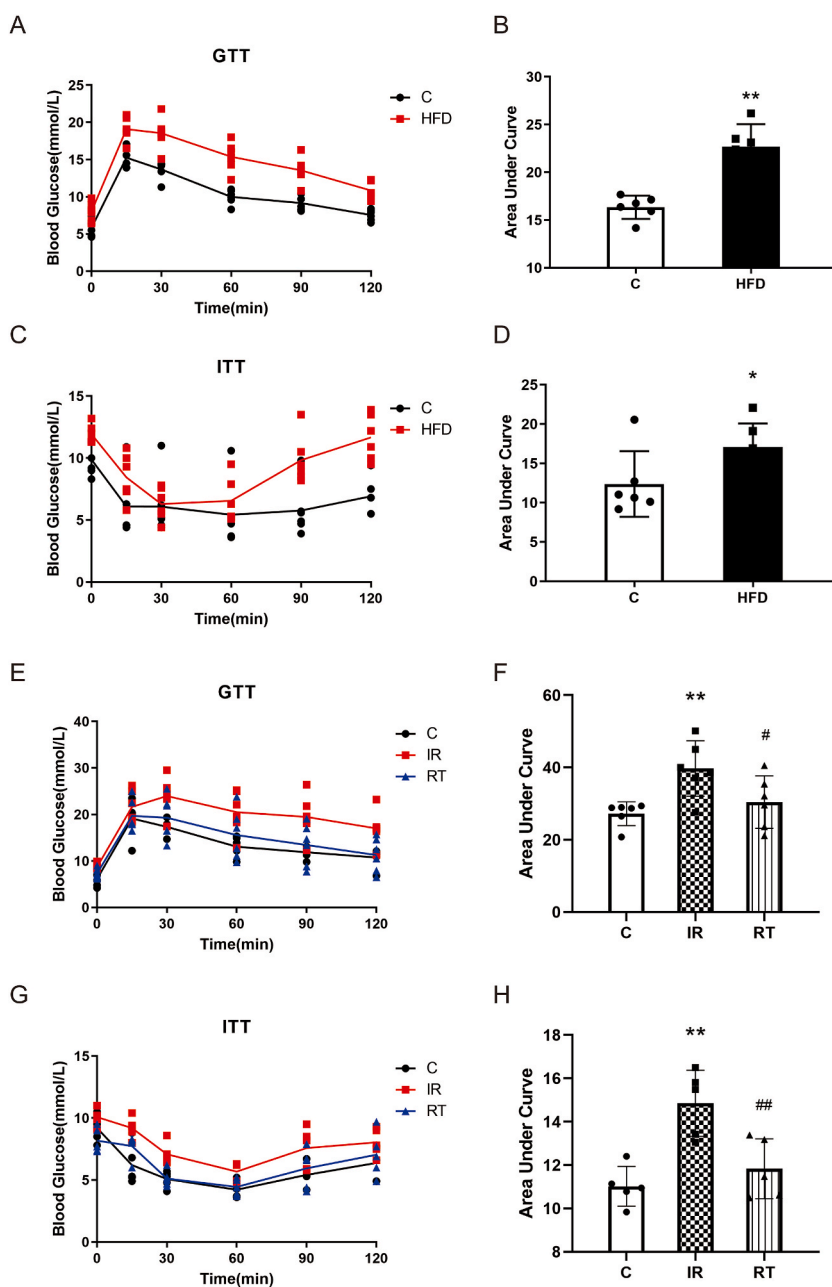
Fig. 1. Effects of HFD and resistance training on insulin sensitivity in mice. (A–D) Glucose tolerance test (GTT) and insulin tolerance test (ITT) curves for the control group and the high-fat diet group (HFD) after 12 weeks of high-fat feeding (n = 5). (E–H) GTT and ITT curves for the control group (C), insulin-resistant group (IR), and resistance training group (RT) after 12 weeks of resistance training intervention (n = 6). Values are presented as mean ± standard deviation (SD). Independent sample *t*-test was used for comparison between HFD and C groups, and one-way analysis of variance (ANOVA) was used for comparison among C, IR, and RT groups. *$P < 0.05$, **$P < 0.01$ vs. control group (C); #$P < 0.05$, ##$P < 0.01$ vs. insulin resistance group (IR).

However, the AUC value of the resistance training group (RT) was significantly lower than that of group IR ($P < 0.05$, Fig. 1E–H).

3.2. Effects of resistance training on plasma lactate of mice with insulin resistance

Compared with group C, plasma lactate level was significantly increased in group IR ($P < 0.01$). Compared with group IR, plasma lactate level was significantly decreased in the RT group ($P < 0.05$, Fig. 2).

3.3. Effects of resistance training on the expression of synaptic plasticity-related proteins in the hippocampus of mice with insulin resistance

To investigate the effects of resistance training on hippocampal plasticity-related protein expression, we examined the expression of NMDAR, postsynaptic density protein 95 (PSD95), and SYN. Compared with group C, the expression levels of PSD95 and SYN in the hippocampus of group IR were significantly decreased ($P < 0.05$). Compared with group IR, the expression levels of NMDAR and SYN in the hippocampus of group RT were significantly increased ($P < 0.05$, Fig. 3).

3.4. Effects of resistance training on PI3K/Akt signaling pathway in the hippocampus of mice with insulin resistance

Given the susceptibility of the hippocampal PI3K/Akt signaling pathway in insulin-resistant mice, we evaluated the phosphorylation and expression levels of PI3K and Akt. In comparison to the control mice, the phosphorylation and total protein levels of PI3K in the hippocampus of insulin-resistant mice did not exhibit significant changes. However, the phosphorylation level of Akt significantly decreased ($P < 0.05$). On the contrary, compared to insulin-resistant mice, the hippocampal phosphorylation and total protein levels of PI3K in the RT group mice showed no significant changes. However, the phosphorylation level of Akt significantly increased ($P < 0.05$, Fig. 4).

3.5. Effects of resistance training on mTOR signaling pathway in the hippocampus of mice with insulin resistance

Because mTORC1 and the synthesis of synaptic plasticity-related proteins go hand in hand, we examined the expression levels of mTOR and Raptor, and the phosphorylation levels of mTOR. Compared with control mice, the phosphorylation level of mTOR in the hippocampus of insulin resistance mice decreased significantly ($P < 0.05$). Compared with insulin-resistant mice, the phosphorylation of mTOR and the expressions of mTOR and Raptor in hippocampus of mice with resistance training increased significantly ($P < 0.05$, Fig. 5A–D). Since PI3K/Akt can further regulate the activity of 4EBP and P70S6 K proteins by regulating mTORC1 to regulate protein synthesis, we have also measured the production and phosphorylation of 4EBP and P70S6 K. Compared with control mice, there was no significant change in the production of P70S6 K in the hippocampus of insulin-resistant mice; the phosphorylation level decreased, and the production of 4EBP2 was significantly increased ($P < 0.05$). Compared with group IR, resistance training didn't significantly change the production and phosphorylation levels of P70S6 K and 4EBP2 in the hippocampus of mice (Fig. 5E–J).

3.6. Effects of resistance training on the expression of glucose transporters (GLUTs) and MCTs in the hippocampus of mice with insulin resistance

PI3K/Akt can also affect the absorption and utilization of glucose by regulating the translocation of GLUTs to the brain cell membrane, which plays an important role in the oxidation and energy supply of cerebral neuronal cells. Therefore, we also measured
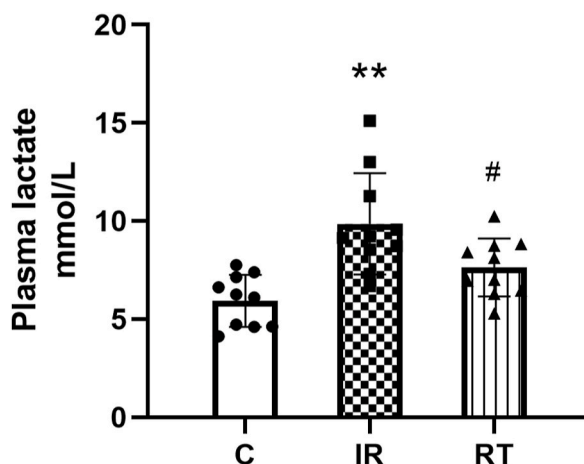
Fig. 2. Effects of resistance training on plasma lactate levels in insulin-resistant mice. All groups (n = 10). Statistical analysis was performed using one-way analysis of variance (ANOVA). **$P < 0.01$ vs. control group (C); #$P < 0.05$ vs. insulin resistance group (IR).
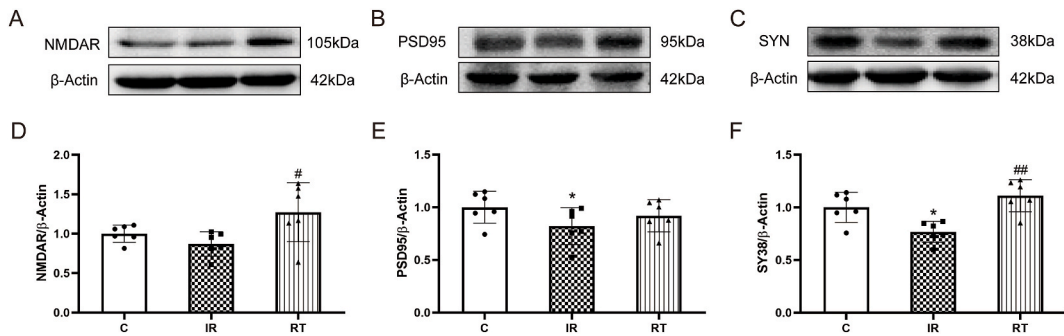
Fig. 3. Effect of resistance training on the expression of synaptic plasticity-related proteins in the hippocampus of insulin-resistant mice. (A–F) Western blot bands and bar graphs for NMDAR, PSD95, and SYN proteins, respectively (n = 6). Statistical analysis was conducted using one-way analysis of variance (ANOVA). *$P < 0.05$ vs. control group (C); #$P < 0.05$ vs. insulin resistance group (IR), #$P < 0.01$ vs. insulin resistance group (IR).
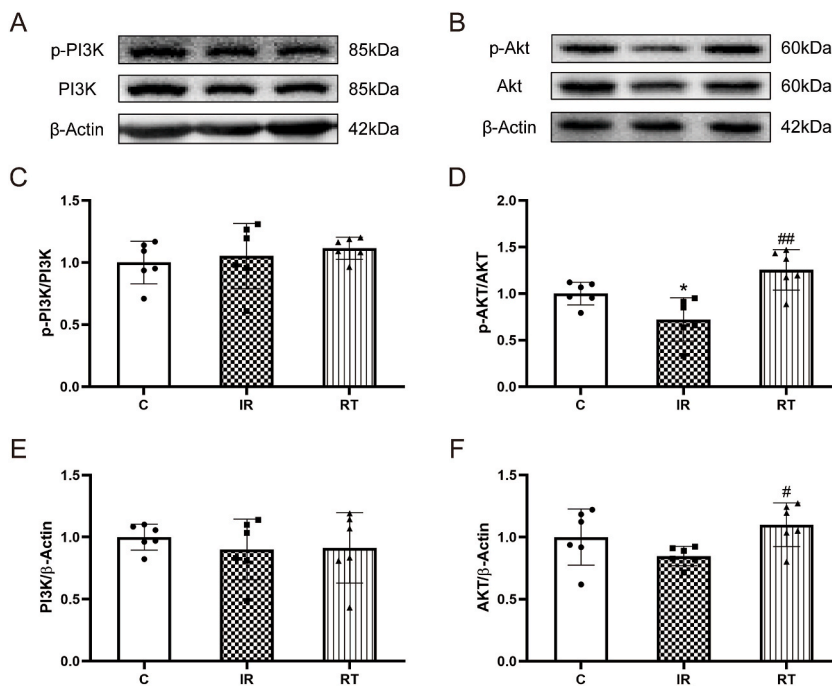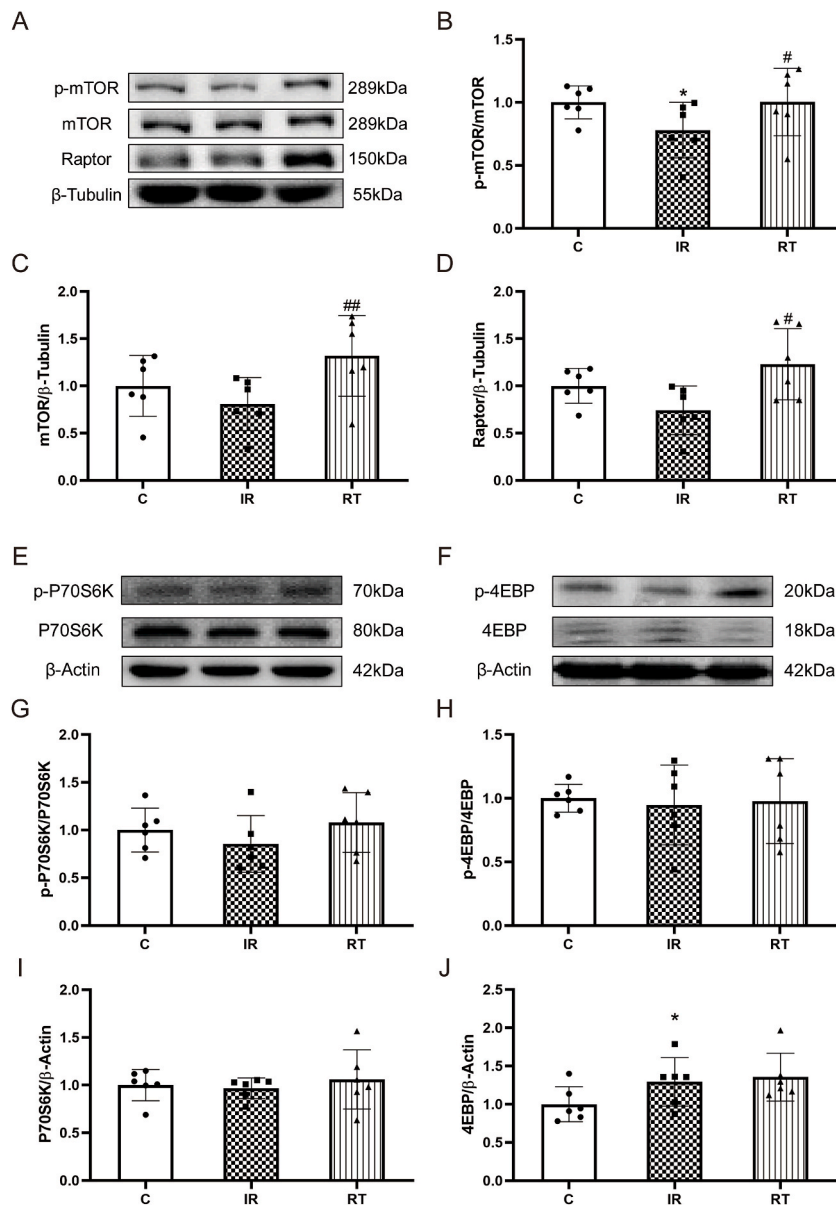

Fig. 4. Effects of resistance training on the PI3K/Akt signaling pathway in the hippocampus of insulin-resistant mice. (A, C, E) Western blot and bar graphs showing PI3K protein expression and phosphorylation levels (n = 6). (B, D, F) Western blot and bar graphs depicting Akt protein expression and phosphorylation levels (n = 6). Statistical analysis was performed using one-way analysis of variance (ANOVA). *$P < 0.05$ vs. control group (C); #$P < 0.05$, ##$P < 0.01$ vs. insulin resistance group (IR).

the production of GLUT1 and GLUT3, the two main forms of GLUTs. Compared with control mice, there was no significant change in the expression of GLUT1 and GLUT3 in the hippocampus of insulin-resistant mice. After 12 weeks of resistance training, there was no significant change in the expression of GLUT1 and GLUT3 in the hippocampus (Fig. 6A–D). However it has been found that, apart from glucose, lactate was also a source of fuel substrate in maintaining normal brain function. The absorption and utilization of lactate need the translocation function of MCTs, so we also measured the expression of MCT1, MCT2, and MCT4. Compared with control mice, the expression of MCT1 and MCT4 in the hippocampus of insulin-resistant mice decreased significantly ($P < 0.05$), and the expression of MCT2 decreased. Resistance training could significantly improve the expression of MCT1, MCT4, and MCT2 ($P < 0.05$, Fig. 6E–J).

4. Discussion

Clinical, epidemiological, and animal studies have demonstrated that diet-induced insulin resistance adversely affects brain function and increases the risk of neurodegenerative disorders such as Alzheimer's disease [21,22]. Clinical, epidemiological, and animal studies have demonstrated that diet-induced insulin resistance adversely affects brain function and increases the risk of

Fig. 5. Effects of resistance training on the mTOR signaling pathway in the hippocampus of insulin-resistant mice. (A–D) Western blot and bar graphs showing mTOR protein expression and phosphorylation, as well as Raptor protein levels (A, n = 6). (E–J) Western blot and bar graphs depicting downstream targets of mTOR, including P70S6 K and 4EBP, and their phosphorylation levels (n = 6). Statistical analysis was performed using one-way analysis of variance (ANOVA). *$P < 0.05$ vs. control group (C); #$P < 0.05$, ##$P < 0.01$ vs. insulin resistance group (IR).

neurodegenerative disorders such as Alzheimer's disease [23]. Proteins such as NMDAR [24], PSD95 [25], and SYN [26,27] play crucial roles in synaptic plasticity, essential for inducing long-term potentiation (LTP) and supporting hippocampus-dependent learning and memory formation [28]. High-fat diet consumption results in diminished PSD95 expression and dendritic spine density in the hippocampus of mice [29], along with decreased SYN expression in the hippocampus of rats [30,31]. Our findings indicate a significant downregulation in the expression of PSD95 and SYN in the hippocampus of mice with diet-induced insulin resistance. Extensive literature supports the beneficial effects of exercise on insulin resistance [32–34], hippocampal neuronal synaptic plasticity [35], and cognitive impairments [36–38]. Although there is substantial evidence for the positive effects of aerobic exercise on cognitive functions [39–41], research on the impacts of resistance training on cognitive functions is less prevalent. Our study provides robust evidence that a 12-week resistance training regimen significantly ameliorates insulin resistance and enhances insulin sensitivity in mice, and it also leads to the upregulated expression of NMDAR and SYN in the hippocampus.

The initial steps in insulin action encompass the phosphorylation of scaffold proteins and the activation of PI3K, which subsequently activates Akt [42]. Akt activation plays a critical role in the insulin-regulated translocation of GLUT4, and diminished
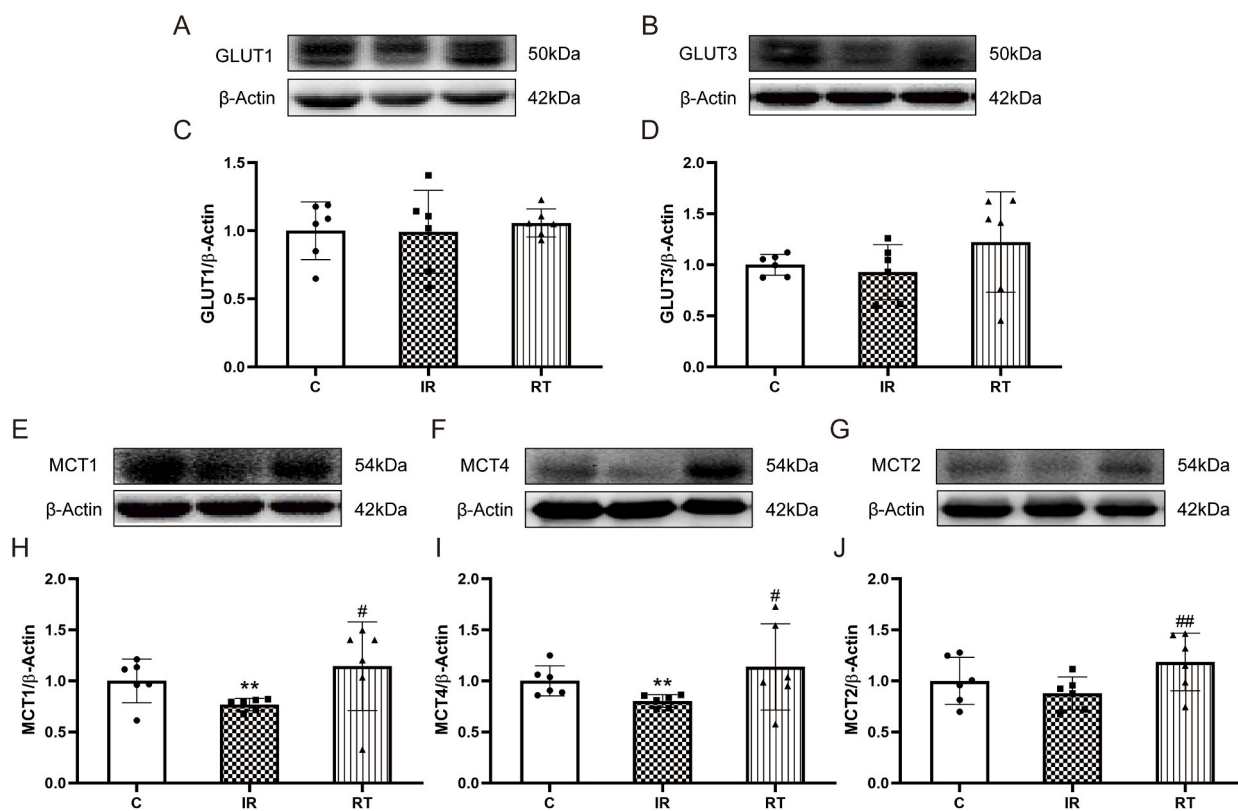
Fig. 6. Effects of resistance training on the expression of GLUTs and MCTs in the hippocampus of insulin-resistant mice. (A–J) Western blot and bar graphs displaying the protein levels of GLUT1, GLUT3, MCT1, MCT4, and MCT2 (n = 6). Statistical analysis was performed using one-way analysis of variance (ANOVA). **$P < 0.01$ vs. control group (C); #$P < 0.05$, ##$P < 0.01$ vs. insulin resistance group (IR).

activation of Akt is a characteristic feature of insulin resistance [43]. In our study, we observed a notable decrease in Akt phosphorylation levels in the hippocampus of insulin-resistant mice, indicating reduced activity of the insulin signaling pathway in this specific brain region, which is suggestive of localized insulin resistance. These findings align with those reported by Pratchayasakul et al. [44], who observed peripheral insulin resistance in Wistar rats following 8 weeks on a high-fat diet. Additionally, they reported a decline in hippocampal neuronal insulin signaling pathway activity after 12 weeks on a high-fat diet, evidenced by reduced phosphorylation levels of the insulin receptor, IRS-1, and Akt in hippocampal slices post-insulin stimulation.

The mTOR pathway, a key downstream target of the PI3K/Akt signaling pathway, combines with the regulatory-associated protein of mTOR (Raptor) and mammalian lethal with SEC13 protein 8 (mLST8) to form mTORC1. This complex plays a vital role in regulating various neuronal processes, including ion channels, receptor pathways, and synaptic plasticity [45,46]. Raptor serves as a bridging molecule, linking mTOR to downstream effectors such as P70S6 K and 4EBP, thus facilitating protein synthesis [47]. Previous research by Maurizio and Jessica [48,49] has highlighted the critical importance of the mTOR-P70S6K/4EBP2 pathway for protein-dependent synaptic plasticity, learning, and memory formation in the hippocampus of rodents. In our study, we assessed the protein expression and phosphorylation levels of mTOR, Raptor, 4EBP2, and P70S6 K in the hippocampus. The results indicated a significant decline in mTORC1 activity and impaired protein synthesis in the hippocampus of insulin-resistant mice, suggesting that inhibition of mTORC1 activity is a key mechanism underlying the reduced expression of synaptic plasticity-related proteins due to decreased activity of the insulin signaling pathway.

Both aerobic exercise and resistance training have been demonstrated to enhance the activity of the insulin signaling pathway in the hippocampus of rodents [50,51]. Aerobic exercise specifically increases the phosphorylation levels of insulin receptors and Akt in the hippocampus of normal mice or rats, leading to elevated expression of synaptic-related proteins such as SYN, PSD95, and β-neurexin [50,52]. In line with these findings, Cassilhas et al. [18] showed that 8 weeks of resistance training significantly boosts Akt phosphorylation in the hippocampus of male Wistar rats. Our findings corroborate these results, indicating that resistance training significantly increases Akt phosphorylation in the hippocampus of insulin-resistant mice, enhancing the activity of the insulin signaling pathway and subsequently increasing mTORC1 activity. This suggests that the benefits of exercise extend beyond normal physiological conditions.

The PI3K/Akt signaling pathway, which is activated in peripheral tissues, facilitates the translocation of GLUT proteins to the cell membrane, enhancing glucose uptake and utilization. Similar signaling mechanisms are active in the hippocampus of rodents [53]. where GLUT1 and GLUT3 are predominantly associated with brain glucose utilization [54,55]. In our study, despite the diabetic

condition, no significant changes were observed in the expression of GLUT1 and GLUT3 in the hippocampus of insulin-resistant mice, aligning with previous findings [56]. Notably, while the overall brain size of diabetic mice was reduced, GLUT1 and GLUT3 expression remained stable. Other research indicates that while GLUT3 expression in the hippocampus of type 2 diabetic rats does not change, a decrease in MCT2 expression has been observed, which may contribute to impaired neuronal lactate uptake and subsequent cognitive dysfunction in type 2 diabetes [57]. Additionally, lactate is increasingly recognized as a supplementary fuel for injured brains, and exogenous lactate supplementation has been shown to alleviate symptoms of acute brain injury [58]. Moreover, the brain can utilize blood lactate as an energy source even under normal conditions, and any disruptions in lactate uptake or transport could further exacerbate the metabolic challenges faced by the brain in the context of diabetes [59].

In a resting state, the brain maintains a dynamic balance between lactate uptake and release. During hypoxia or intense physical activity, blood lactate levels rise, and the brain's lactate uptake surpasses its release, suggesting lactate as a viable alternative energy substrate for the brain [60]. Studies have shown that during hypoglycemia, lactate infusion can sustain normal brain evoked potentials and supply up to 60 % of the brain's energy needs, underscoring lactate's critical role in brain function [61]. Lactate transport from the periphery to the brain or from astrocytes to neurons depends on monocarboxylate transporters (MCTs), particularly MCT1, MCT2, and MCT4, which are integral to brain lactate transport [11]. In type 2 diabetic rats, research has indicated an increase in hippocampal glycogen levels coupled with a decrease in MCT expression, leading to impaired memory function [57]. This suggests that diminished lactate uptake due to reduced MCT expression could contribute to spatial memory deficits. Experiments with neuron-specific MCT2 knockout rats have confirmed MCT2's crucial role in maintaining synaptic transmission through lactate uptake [62]. Further, recent studies have demonstrated that brain lactate not only acts as an energy source but also modulates the activity of adenosine mono-phosphate (AMP)-activated protein kinase (AMPK), an energy sensor protein regulated by the ratio of AMP to adenosine triphosphate (ATP). An increase in AMP/ATP levels activates AMPK, which in turn can inhibit mTORC1 activity, subsequently reducing protein synthesis [12]. Thus, alterations in MCT expression could directly impact synaptic plasticity-related protein expression through changes in energy availability. In our research, insulin-resistant mice exhibited a significant increase in plasma lactate levels along with reduced expression of hippocampal MCT1 and MCT4. This suggests that chronic high-fat diet-induced insulin resistance not only impairs the body's oxidative capacity and elevates lactate production but also hinders the brain's ability to uptake lactate effectively from the periphery and astrocytes. As a result, neuronal lactate uptake is diminished, leading to lower neuronal lactate levels. Given the essential role of brain lactate metabolism in sustaining normal brain function [61], this could be a pivotal mechanism contributing to the observed decline in synaptic plasticity-related protein expression in the hippocampus of insulin-resistant mice in our study.

Previous research on the impact of exercise on blood lactate has predominantly concentrated on aerobic exercise. Studies have shown that combined aerobic and resistance training effectively improves blood lactate levels in individuals with type 2 diabetes and enhances insulin sensitivity, indicating a potential relationship between lactate reduction and improved insulin response [63]. Additionally, moderate-intensity treadmill exercise has been found to mitigate the decrease in MCT2 expression in the hippocampus of diabetic animal models, resulting in enhanced spatial memory [57]. This effect is likely due to the upregulation of hippocampal glycogen levels and MCT2 expression, which facilitates the transport of glycogen-derived lactate to neurons, thereby increasing the energy supply. However, the long-term effects of single-mode resistance training on blood lactate levels have not been extensively explored. Our study contributes to the understanding of this aspect by demonstrating that resistance training effectively reduces blood lactate levels and improves insulin resistance. Moreover, the impact of resistance training on hippocampal MCT expression has been largely unexplored in existing literature. Our findings, which show an upregulation of hippocampal MCT expression in insulin-resistant mice following exercise, suggest for the first time that resistance training can effectively counteract the excessive elevation of blood lactate levels in these mice. This leads to increased expression of MCT1, MCT4, and MCT2 in the hippocampus, enhancing lactate transport to the brain and its uptake by neurons. These processes may be crucial in upregulating synaptic plasticity-related protein expression in the hippocampus of insulin-resistant mice.

Traditionally, lactate has been considered a metabolic by-product and a major contributor to exercise-induced fatigue. However, a growing body of evidence now suggests that lactate can also serve as a viable energy substrate [64–66]. Our study adds to this perspective by suggesting that enhanced lactate transport to the brain may represent another significant mechanism through which exercise promotes brain health in individuals with insulin resistance. This novel viewpoint offers new avenues for exploring the mechanisms underlying the beneficial effects of exercise on brain health, with a specific focus on lactate metabolism.

5. Conclusion

This study investigates changes in the PI3K/Akt-related pathway and the expression of synaptic plasticity-related proteins in the hippocampus of mice with peripheral insulin resistance. These findings offer crucial insights into the theoretical underpinnings of cognitive decline associated with insulin resistance and type 2 diabetes. Moreover, the long-term effects of resistance exercise on cognitive enhancement in models of insulin resistance have not been thoroughly examined, particularly concerning brain energy metabolism. Future research should focus on exploring this aspect to deepen our understanding of exercise's beneficial effects (Fig. 7).
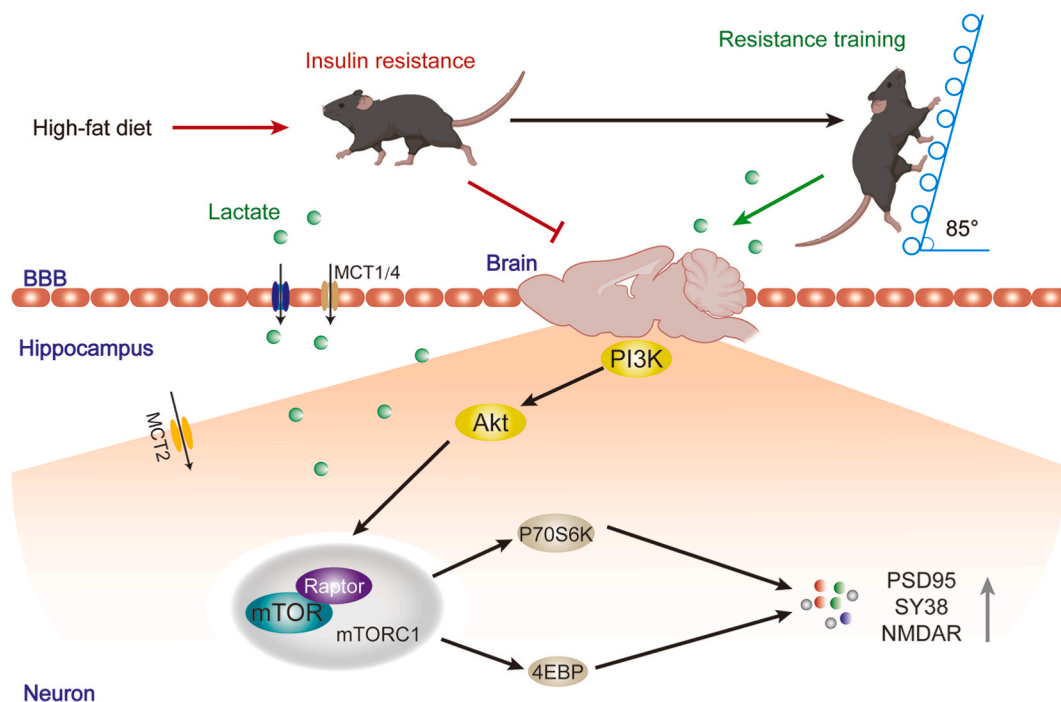
Fig. 7. Graphical abstract. Effects of long-term resistance training on insulin resistance and expression of central synaptic plasticity-related proteins.

Data availability

The datasets generated or analyzed during this study are available from the corresponding author on reasonable request.

Ethics declaration

The experimental design and animal use were set by the Institutional Animal Ethics Committee (IAEC) guidelines and approved by the Scientific Research Ethics Committee of Shanghai University of Sport (2015013).

CRediT authorship contribution statement

Xuepeng Bian: Writing – review & editing, Writing – original draft, Validation, Methodology, Investigation, Formal analysis, Data curation. **Mingming Li:** Writing – review & editing. **Shujie Lou:** Writing – review & editing, Supervision.

Declaration of competing interest

The authors declare that there are no conflicts of interest regarding the publication of this paper. There have been no financial, consultative, institutional, or other relationships that could lead to a perceived conflict of interest in relation to the research and/or publication of this manuscript.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.heliyon.2024.e34425.

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