Higher voluntary wheel running activity following endurance exercise due to oral taurine administration in mice

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Abstract The aim of this study was to examine the effects of oral taurine (2-aminoethanesulfonic acid) administration on the amount of voluntary wheel running as an indicator of recovery from endurance exercise-induced fatigue in ICR mice. We orally administered a single dose of taurine (0.5 mg/g body weight) or physiological saline immediately after treadmill running at 25 m/min for 90 min. After administration, we placed mice in cages with a running wheel and allowed them to run freely in the wheel with free access to food. In the saline-treated group, exercise significantly decreased the amount of voluntary wheel running compared to the non-exercised mice (p < 0.01), while exercise did not decrease the amount of voluntary wheel running in the taurine-treated group. Significant effects of post-exercise taurine administration on voluntary wheel running during 6 h were found (p < 0.05). The 30-min running distance was significantly higher in the taurine-treated group than in the saline-treated group at 1-1.5 h after treadmill exercise (p < 0.05). Blood glucose and liver and skeletal muscle glycogen concentrations after treadmill exercise were similar in both groups at all times. Total food consumption during 6 h of voluntary wheel running showed no difference between the two groups. The ratio of the total running distance to total food consumption was significantly higher in the taurine-treated group than in the saline-treated group (p < 0.05). Our results show that oral taurine administration after endurance exercise increased the amount of voluntary wheel running. Taurine administration may have a positive effect on recovery from endurance exercise-induced fatigue.

Keywords: taurine, endurance exercise, voluntary wheel running, food consumption, recovery

Introduction

Although it is very difficult to define what fatigue is, fatigue can be defined as a state of deterioration in the homeostatic body condition. Endurance exercise can be a potent stimulus that induces fatigue, which is caused by many factors, including decreased muscle and liver glycogen concentrations, increased muscle sodium concentrations, decreased muscle potassium concentrations, increased body temperature, dehydration, etc. During recovery from endurance exercise, the inner body condition gradually returns to a normal homeostatic state in a few hours; however, it may take several days to recover from muscle injury, increased cytokine response etc. It is important to investigate factors related to faster recovery from fatigue caused by exercise. However, there are not effective methods to investigate recovery after exercise-induced fatigue in animal models. Some groups have measured the amount of voluntary wheel running in mice during the post-exercise phase to investigate recovery from exercise-induced fatigue. In these studies, the distance of voluntary wheel running was significantly decreased by exercise, and then gradually increased to the pre-exercise level. It can be assumed that mice that are able to recover from exercise faster can run a longer distance in a wheel. Thus, voluntary wheel running seems to be a good model for the examination of recovery from exercise. In many studies, voluntary wheel running was used as an indicator of recovery from muscle inflammation. The activity of voluntary wheel running is thought to be affected not only by inflammation, but also by many central and peripheral factors. One possible factor affecting voluntary wheel running activity is the capacity of energy metabolism. Dumke et al. reported that insulin-stimulated glucose uptake in skeletal muscle was significantly higher in mice genetically selected for high wheel running activity. In addition, a positive correlation between running distance and food consumption has been demonstrated in mice, reflecting a higher energy demand for wheel running activity. Because endurance exercise depletes the glycogen storage in liver and skeletal muscle, higher voluntary wheel running activity during the post-exercise phase may reflect an improvement in metabolism in the form of faster glycogen resynthesis and/or higher mobilization of fatty acids.

In this study, we focused on the effects of taurine...
(2-aminoethanesulfonic acid) administration on voluntary running activity after endurance exercise. Taurine, which is present in high concentrations in excitable tissues such as neurons and cardiac and skeletal muscles, can support the contractile properties of skeletal muscle\(^9\)\(^{-11}\). It is suggested that taurine can act as a buffer to maintain osmolality and a homeostatic environment of the body partly because it can be easily dissolved\(^9\). Taurine cannot be used as an energy substrate, and it does not regulate enzyme activity or gene expression directly\(^9\). It has been suggested that taurine administration improves energy metabolism, though the mechanisms by which this occurs have not been clarified. For example, taurine administration before exercise increases fatty acid oxidation\(^2\) and can maintain a higher blood glucose concentration during prolonged exercise\(^13\)\(^{14}\). It has been shown that endurance exercise decreases the taurine content in many tissues including skeletal muscle in both rodents and humans, possibly due to the excretion of taurine from skeletal muscle into the plasma and urine\(^15\)\(^{-18}\). Taurine administration after exercise-induced taurine depletion may have some positive effects on energy metabolism during the recovery phase.

We investigated whether taurine administration after exercise can promote recovery from exercise by measuring voluntary wheel running activity after a single session of endurance exercise at 25 m/min for 90 min, which can be expected to decrease taurine content in the skeletal muscles of mice based on previous rat studies\(^17\)\(^{18}\). We also measured the concentrations of metabolic substrates in skeletal muscle, liver, and blood to investigate the effects of taurine administration on substrate metabolism following endurance exercise.

**Materials and Methods**

**Animals.** Six-week-old male ICR mice were purchased from CLEA Japan, Inc. (Tokyo, Japan). Mice were housed in a room maintained at 23°C with three mice per cage, and were acclimatized for 1 week. Mice were given free access to a standard chow (MF; Oriental Yeast Co., Ltd., Tokyo, Japan). Other studies using the same chow found access to a standard chow (MF; Oriental Yeast Co., Ltd., Tokyo, Japan). Other studies using the same chow found.

**Experimental methods.** All experimental treatments were performed in the dark phase (9AM-9PM) when mice were active. One week before the experiment, the mice were placed in voluntary activity wheel cages (22 × 9 × 8 cm) with a running wheel (0.2 m diameter, CL-4579-2; CLEA Japan, Inc.) for 3 days. Mice were allowed to run freely in the wheel and were fed *ad libitum*. The number of wheel revolutions in both directions was counted. The running distance was determined from the number of revolutions. Mice were then divided into a taurine-treated group (n = 5 to 7) and a physiological saline-treated group (n = 5 to 7) with similar mean body weights and mean running distances during the 3 days of voluntary wheel running. Three days before the treadmill exercise, all mice were familiarized with treadmill exercise at a speed of 25 m/min for 5 min. In all experiments, mice in the taurine-treated group were orally administered 0.5 mg/g body weight of taurine dissolved in physiological saline (0.9%) using a sonde.

On the day of the experiment, after mice were fasted for 4 h to avoid a postprandial state, they ran on the treadmill at 25 m/min for 90 min. Immediately after treadmill running, mice were orally administered taurine or physiological saline. All mice were then placed in voluntary activity wheel cages with a running wheel. Mice were allowed to run freely in the wheel and were fed *ad libitum*. The running distance of the mice was counted every 30 min. Mice were sacrificed by cervical dislocation at 0, 3, or 6 h from the start of the voluntary wheel running. Blood samples were taken immediately from the open chest. Plasma, liver, and skeletal muscles of the lower hind limb were frozen in liquid nitrogen and kept at -80°C until further analysis. The total amount of food consumption was determined by weighing the chow at the start and end of the experiment.

To measure the concentrations of metabolic substrates during the resting and fed state (baseline), mice (n = 6) were sacrificed at rest by cervical dislocation. To measure the amount of running wheel activity without advanced endurance running, some mice (n = 7) were placed in voluntary wheel cages for 6 h.

**Analytical methods.** Blood glucose concentrations were measured using an auto analyzer (Glutest Ace; Arkray Inc., Kyoto, Japan). Plasma free fatty acid (FFA) concentrations were measured using a kit (Wako NEFA C test kit; Wako Pure Chemical Industries, Ltd., Osaka, Japan). Gastrocnemius muscle and liver glycogen concentrations were measured using the phenol-sulfuric acid method\(^9\).

**Statistical analysis.** All values are expressed as mean ± SE. We used Prism 5 software (Graph Pad Software, San Diego, CA) for the analyses. The changes in each metabolic substrate during the experiment were compared by one-way analysis of variance (ANOVA). When differences were found to be significant, comparisons were made using Tukey’s post hoc (HSD) test. The total running distance was analyzed by two-way (treatment × time) repeated-measures ANOVA to determine differences in running distance between the taurine-treated and saline-treated groups, and between exercised and non-exercised mice. When differences were detected, comparisons were made using the Bonferroni test. We used a two-way (treatment
× time) nonrepeated-measures ANOVA to determine differences in metabolic substrate concentrations in tissues between the taurine-treated and saline-treated groups. The difference in the ratio of total running distance to total food consumption between the two groups was analyzed by an unpaired t-test. Statistical significance was set at p < 0.05.

Results

Voluntary wheel running activity. In the saline-treated group, 90 min of endurance running at 25 m/min significantly reduced the amount of voluntary wheel running compared with mice that did not perform endurance exercise (p < 0.01) (Fig. 1A). No significant difference in the amount of voluntary wheel running was observed between exercised and non-exercised mice in the taurine-treated group (Fig. 1B). There was no significant difference in the amount of voluntary wheel running between the non-exercised saline-treated group and non-exercised taurine-treated group (Fig. 1C). The total amount of voluntary wheel running for 6 h after treadmill running was significantly higher in the exercised taurine-treated group than in the exercised saline-treated group (p < 0.05) (Fig. 1D). The voluntary 30-min wheel running activity at 1 - 1.5 h after treadmill running was higher in the exercised taurine-treated group than in the exercised saline-treated group (p < 0.05). There was a main effect of taurine administration on the amount of voluntary wheel running (p < 0.01) (Fig. 1E).

Metabolic substrate contents in blood and tissues. Endurance running at 25 m/min for 90 min decreased the concentrations of blood glucose (8.6 ± 0.7 mmol/l at resting state vs. 2.5 ± 0.2 mmol/l at the end of exercise, p < 0.01), gastrocnemius muscle glycogen (2.4 ± 0.4 mg/g at resting state vs. 0.8 ± 0.3 mg/g at the end of exercise, p < 0.05), and liver glycogen (26.7 ± 1.5 mg/g at resting state vs. 5.5 ± 2.5 mg/g at the end of exercise, p < 0.01). After 3 h of voluntary wheel running, there were no significant differences in the metabolic substrate concentrations in tissues between the taurine-treated and saline-treated groups.
differences in the blood glucose and liver glycogen concentrations compared with baseline in both groups (Fig. 2A and B). Furthermore, after 6 h of voluntary wheel running, liver glycogen concentration was significantly increased from that at baseline and at 3 h (p < 0.01) (Fig. 2B). Glycogen concentration in the gastrocnemius muscle did not reach the baseline level after 3 h of voluntary wheel running, but reached the baseline level during 6 h of voluntary wheel running in both groups (p < 0.05) (Fig. 2C). Plasma FFA concentration was significantly increased by endurance exercise (p < 0.05); there were no significant differences in the plasma FFA concentration between the baseline level and after 3 and 6 h of voluntary wheel running (Fig. 2D). No significant differences were found in the concentrations of metabolic substrates in blood and tissues between the taurine-treated and saline-treated groups at any time.

**Relationship between total running distance and food consumption.** There were no significant differences in body weights at the start of the voluntary wheel running phase between the two groups. There was also no significant difference in total food consumption during 6 h of recovery between the two groups (52.8 ± 5.6 mg/g body weight in the taurine-treated group vs. 47.2 ± 6.6 mg/g body weight in the saline-treated group). The ratio of total distance to total food consumption was significantly higher in the taurine-treated group than in the saline-treated group (p < 0.05) (Fig. 3).

**Discussion**

Endurance running at 25 m/min for 90 min induced hypoglycemia and depletion of muscle and liver glycogen in mice. We used voluntary wheel running as an indicator of recovery from exercise-induced fatigue in mice that had ingested saline or taurine immediately after the exercise. The amount of voluntary wheel running was significantly decreased by endurance exercise in the saline-treated group, while it was not reduced by endurance exercise in the taurine-treated group. We also found that the total voluntary wheel running distance after endurance exercise was significantly higher in the taurine-treated group than in the saline-treated group. Our results suggest that ad-

![Fig. 2](image_url)  
(A) Blood glucose concentration, (B) liver and (C) gastrocnemius muscle glycogen concentrations, and (D) the plasma free fatty acid concentration in mice orally administered taurine (white bar, n = 7) or saline (black bar, n = 7) immediately after a single session of treadmill running at 25 m/min for 90 min. For comparison, we also measured the concentration of each metabolite in tissues from mice kept at rest (hatched, baseline, n = 6) and mice sacrificed immediately after endurance exercise (hatched, 0 h, n = 5). Values are means ± SE. The changes in each metabolic substrate during the experiment were compared by one-way ANOVA. ‡p < 0.05 and ‡‡p < 0.01 vs. baseline; §§p < 0.01 vs. 3 h. The differences between the taurine-treated and saline-treated groups were compared by two-way ANOVA.

![Fig. 3](image_url)  
Ratio of total running distance to total food consumption (m/g) in mice orally administered taurine (white, n = 12) or saline (black, n = 14). Values are means ± SE. The unpaired t-test was used for statistical analysis. *p < 0.05 vs. the saline-treated group.
ministration of taurine has some positive effects on recovery from fatigue induced by endurance exercise, though further studies are necessary to elucidate the mechanisms of these taurine effects on recovery from exercise-induced fatigue.

In rat studies, an increased taurine concentration in skeletal muscle was observed after oral taurine administration for 1, 7, and 14 days without exercise, while the taurine concentration in skeletal muscle did not increase in humans receiving taurine without exercise for 7 days. In rats, orally ingested taurine was rapidly absorbed from the gastrointestinal tract and detected in the blood and many tissues including the liver and urinary bladder within 10 min after administration. In addition, taurine was detected in skeletal muscle 15 min after intravenous injection in rats. Therefore, we can expect that orally administered taurine will appear in the blood and reach the skeletal muscles within 1 h in mice. During exercise, skeletal muscle loses various ions and other solutes including taurine to compensate for increases in many osmotically active molecules as a result of enhanced energy metabolism. Indeed, skeletal muscle taurine concentration in rats was significantly lower after prolonged treadmill exercise (25 m/min for 60 or 100 min) than at rest. In this study, the taurine concentration in skeletal muscle was expected to be decreased by treadmill exercise at 25 m/min for 90 min. A previous study showed that oral taurine administration increased skeletal muscle taurine concentration in aged rats, which had lower taurine concentration compared with young rats. Collectively, these findings show that orally administered taurine should be transported into skeletal muscle cells after the taurine content is decreased by exercise in mice. Miyazaki et al. investigated the effects of the optimal and effective doses of pre-exercise administered taurine on endurance capacity in rats. They showed that taurine administration at 500 mg/kg body weight was the most effective dose for exercise time prolongation compared to 20 or 100 mg/kg body weight. Therefore, we treated the mice in the present study with orally administered taurine at a dose of 0.5 mg/g body weight after exercise and investigated the effect of taurine administration during the post-exercise recovery phase. Taurine seems able to support various physiological processes. Previous studies have suggested that taurine has some effects on the contractile properties of skeletal muscle, including the release and uptake of Ca²⁺ by the sarcoplasmic reticulum in the excitation-contraction coupling process, prevention of peroxidation of plasma membranes, and recovery from exercise-induced cellular membrane damage. In addition, taurine transporter knockout (taut−/−) mice, showing reduced taurine concentration in skeletal muscles and tissues, had a significantly lower endurance capacity (>80%) compared to wild-type mice. Although the exact effect of taurine administration on skeletal muscle function has not yet been elucidated, it is possible that taurine administration results in faster recovery of cellular osmolality or other functions of skeletal muscle after endurance exercise, leading to higher wheel running activity.

In the present study, glycogen concentrations in liver and muscle were significantly lower than that at baseline at the end of the endurance exercise at 25 m/min for 90 min. The liver glycogen concentration returned to the baseline level at 3 h of recovery with voluntary wheel running, but the muscle glycogen concentration was still lower than the baseline level at 3 h. The muscle glycogen concentration returned to the baseline level at 6 h of recovery with voluntary wheel running. These results suggest that in this study, the main fuel for voluntary wheel running was the endogenous lipid storage pool, particularly in the early recovery phase. A recent study reported that taurine treatment increased the catalytic activity of cAMP-activated protein kinase A in white adipocyte tissue cells. This kinase is important for the activation of hormone-sensitive lipase, which plays an important role in increasing lipolysis in both adipocytes and skeletal muscle. In addition, a single oral dose of taurine before exercise reportedly increased fat oxidation during prolonged exercise in humans. The higher ratio of the total voluntary running distance to total food consumption in the taurine-treated group after exercise-induced glycogen depletion might be associated with increased mobilization of endogenous lipids by taurine treatment. Another possible explanation of why post-exercise taurine administration increases voluntary wheel running is the enhancement of glucose uptake and/or glycogen synthesis, as evidenced by previous reports showing that taurine administration improved insulin sensitivity in many tissues and enhanced glycogen synthesis in the liver. In this study, we found no differences in either the glycogen concentrations in skeletal muscle and liver or the plasma FFA concentration between the two groups during voluntary wheel running. However, it is possible that a higher voluntary wheel activity offsets the effect of taurine administration because of a higher energy demand. Further study is necessary to investigate the effects of post-exercise taurine administration on glycogen synthesis and lipid utilization in mice at rest during the recovery phase. Activation of glycogen resynthesis during the post-exercise phase is important for faster recovery from endurance exercise. Some studies showed that protein or amino acids intake with carbohydrate can enhance glycogen synthesis in skeletal muscle after endurance exercise, possibly due to increased plasma insulin concentration. In addition, previous studies of rats and horses indicated the effectiveness of post-exercise administration of acetate, which can be converted to acetyl-CoA and then used as a substrate for the TCA cycle. Increased utilization of energy substrates other than glucose and glycogen seems to be concomitant with increased glycogen resynthesis. Although the mechanism of action of taurine has not yet
been clarified, taurine administration may improve energy metabolism by enhancing glucose uptake\(^{12,34}\) and/or lipid mobilization\(^{12,31}\). In addition to its possible effects on energy metabolism, taurine administration may have positive effects on the contractile properties of skeletal muscle, as mentioned above. Thus, it is also possible that taurine administration after endurance exercise leads to faster recovery of the contractile properties of skeletal muscle.

Conclusions

We examined the effect of taurine administration, after endurance exercise (25 m/min for 90 min), on the amount of voluntary wheel running as an indicator of recovery from fatigue. In the saline-treated group, the amount of voluntary running wheel activity was significantly lower after endurance exercise compared to the non-exercised condition, while exercise did not decrease the amount of voluntary wheel running in the taurine-treated group. We found significant effects of taurine supplementation on the total voluntary wheel running activity during recovery from endurance exercise. These results suggest that taurine administration has some favorable effects on recovery from fatigue caused by endurance running.

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References


