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Anti-Fatigue, Antioxidation, and Anti-Inflammatory Effects of Eucalyptus Oil Aromatherapy in Swimming-Exercised Rats

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Abstract

Eucalyptus globulus **possesses important pharmacological activities, including antioxidant and anti-inflammatory effects. We investigated the anti-fatigue, antioxidant, and anti-inflammatory effects of eucalyptus essential oil after swimming exercise using an animal model. Male Sprague–Dawley rats were administered eucalyptus oil (200 μL/h) daily *via* inhalation (15 min), and anti-fatigue effects were assessed following eucalyptus essential oil administration for 2 or 4 weeks when forced to swim until exhaustion while carrying ~5% body weight-equivalent. To assess antioxidant and anti-inflammatory effects, control and oil-treated groups were subjected to swimming, which was intensified from 90 min to 120 min daily over 4 weeks, with non-swimming groups included as controls. The 2- and 4-week-treated rats increased their swimming-to-exhaustion time by 46 s and 111 s, respectively. Additionally, lactate (LA), creatine kinase (CK), and lactate dehydrogenase (LDH) activities increased significantly in the non-treated swimming relative to levels observed in the non-swimming groups ($P < 0.05$); however, no significant differences in these markers were observed between the treated groups. The anti-fatigue effects were related to LA clearance and reduced LDH and CK concentrations. Moreover, compared to the corresponding levels in the non-swimmers, the non-treated swimmers showed markedly elevated levels of liver malondialdehyde (MDA), xanthine oxidase (XO), and other factors, but significantly decreased ($P < 0.05$) glutathione (GSH) concentrations. However, compared with that of the non-swimmer group, the treated swimming group showed no significant changes in these levels ($P > 0.05$), suggesting stable XO and MDA production and maintenance of GSH levels. These results suggested that eucalyptus oil aromatherapy increased rat swimming performance and antioxidant capacity and decreased oxidative damage and inflammatory reactions in tissues, indicating good anti-fatigue, antioxidant, and anti-inflammatory effects after high-intensity endurance exercise.**

Key Words: 1,8-cineole, anti-fatigue, anti-inflammation, antioxidation, *Eucalyptus globulus*

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Introduction

Fatigue caused by high-intensity endurance exercise can affect exercise performance and even lead to sports injuries, with greater risk of such negative effects increasing along with exercise intensity. When the body is at rest, the electron-transport chain transforms ~2% to 5% of the oxygen molecules into oxygen free radicals (12). During strenuous exercise, the body requires an intake of ~10- to 20-times the amount of oxygen consumed at rest, and the oxygen content in the muscles increases 100- to 200-fold (8). Therefore, an excess of reactive oxygen species (ROS) is produced, resulting in oxidative damage. Additionally, strenuous exercise induces numerous immune-system proinflammatory reactions similar to those induced by injury or burns (28). To counteract these phenomena caused by strenuous exercise, the search for anti-fatigue, antioxidative, and anti-inflammatory therapies has become increasingly important. Aromatherapy is a common form of complementary treatment used in Europe and the United States to restore health (13) and involves the delivery of essential oils extracted from aromatic plants into the body through the skin, respiratory tract, or digestive tract by various means, such as massaging, bathing, incense inhalation, and ingestion.

Numerous types of essential oils exist with varying compositions and effects. One such example is *Eucalyptus globulus*, which contains 1,8-cineole (eucalyptol) that has important pharmacological activities, including antioxidant and anti-inflammatory effects (6). The molecular formula of 1,8-cineole, a monoterpene, is $C_{10}H_{18}O$, and its molecular weight is 154.24 g/mol. This compound can significantly improve respiratory function and is, therefore, often used to treat respiratory diseases, such as dyspnea, bronchitis, rhinitis, and asthma (30). In patients with chronic bronchitis, it can exert antioxidant effects on lipid peroxides by inducing catalase (CAT) activity in red blood cells (35). Additionally, 1,8-cineole can significantly inhibit tumor necrosis factor-α and interleukin-1β *in vitro*. Therefore, it was hypothesized that 1,8-cineole might exert similar effects on cytokines (20). Because the mechanisms underlying inflammation mainly include generation of ROS, 1,8-cineole can scavenge hydroxyl radicals ($.OH^-$) and alleviate inflammatory symptoms (27). However, current research on the use of essential oil aromatherapy in sports training is scarce and mostly emphasizes the potential calming, relaxing, or stimulatory effects of essential oils during exercise (14). Therefore, the potential anti-fatigue, antioxidant, and anti-inflammatory effects of essential oils during strenuous exercise are worth investigating.

Materials and Methods

Experimental Animals

We purchased 64 male, 8-week-old, Sprague–Dawley rats weighing 250 g to 300 g from the National Laboratory Animal Center (Taipei, Taiwan). After a 1-week environmental acclimatization, the rats were subjected to 10 min of swimming daily for 3 days before initiation of the experiments. All experimental procedures were conducted in accordance with relevant guidelines and approved by the Ethics Committee of Da-Yeh University (protocol number: DYU102004).

Materials

The 100% pure eucalyptus oil distilled from *E. globulus* was obtained from Laboratoire Vie Arome (Graveson, France), with a eucalyptol content of 83.036% according to gas chromatography mass spectrometry analysis.

Inhalation and Dose Determination

A literature search was performed to identify a method for evaluating the adequate dose of essential oils (36). Aromatherapy was administered *via* spray inhalation. An anion atomizer and the principle of electronic oscillation were used to apply high-frequency oscillations to break up the essential oil. This process produced a mist comprising oil particles with diameters ranging from 0.5 μm to 6 μm, which were delivered to the rats by inhalation.

To assess the tolerable dose range of eucalyptus oil for the rats, they were divided into the following experimental groups and treated as indicated: control (untreated), and low-, medium-, and high-dose (100, 200, and 400 μL/h, respectively) groups (n = 6/group). Each group of rats from the same cage was placed in a plastic box (length: 60 cm; width: 40 cm; height: 40 cm). The rats were exposed to essential oil inhalation for 15 min, after which they were subjected to a weight-bearing forced swimming exercise to observe the number of times each rat extended its head, sank below the water, and exhibited other such behaviors. Total swim time was evaluated to assess the optimal dose of essential oil.

Weight-Bearing Forced Swim Test and Dosage Determination

As described in a previous study (24), a metal wire (~5% of the body weight) was tied to the body of each rat, and they were subsequently placed in a transparent glass tank (length: 90 cm; width: 30 cm; height: 60 cm) containing water maintained at a

temperature ranging from 27°C to 29°C and allowed to swim freely. The rats were continuously monitored by video recording while swimming, and the swim time was measured. For the weight-bearing forced swim, control and low-, medium-, and high-dose groups swam for 511 ± 58.03 s, 503.2 ± 61.86 s, 545 ± 82.61 s, and 361.81 ± 50.20 s, respectively. These results showed that the weight-bearing swim time of the high-dose group decreased significantly ($P < 0.05$). Moreover, the rats shook their heads and tilted their limbs while swimming, indicating that the high essential oil dose might have been excessive, resulting in dizziness (25). No significant differences in the weight-bearing swim time were observed among the remaining three groups ($P > 0.05$), although the medium-dose group exhibited increased exercise performance. Based on these results, the medium dose (200 μL/h) was selected for use in this study.

Anti-fatigue, Antioxidation, and Anti-inflammatory Testing of Eucalyptus Essential Oil Anti-fatigue testing.

The rats were randomly divided into four experimental groups. The control group was not subjected to swimming exercise (C), whereas the remaining three groups [*i.e.*, without inhalation of oil (0 Oil) and with oil inhalation for 2 weeks (2w + Oil) and 4 weeks (4w + Oil)] were subjected to swimming exercise. Rats from the latter two groups were administered eucalyptus oil *via* inhalation (15 min) once daily at a dose of 200 μL/h for 2 and 4 weeks. At the end of the 4-week experimental period, food was withdrawn from all rats for 6 h, after which they were subjected to the weight-bearing forced swim test to measure the swim time in order to assess anti-fatigue exercise performance. The procedure for the weight-bearing forced swim test was the same as that used during establishment of the adequate dose of essential oil. The swim time was determined as the time from entering the water to the point at which the rat could not continue swimming, sank for 10 s, and failed to return to the water surface. The rats were then immediately euthanized using CO_2 asphyxiation, and blood was collected from the aorta.

Antioxidation and anti-inflammatory testing. The rats were randomly divided into four experimental groups. Of these, two groups were not subjected to swimming [*i.e.*, the untreated control group (C) and the group treated with essential oil (Oil)], whereas the other two groups were subjected to swimming either without (S) or with (S + Oil) essential oil administration. The rats from the latter two groups were subjected to 90 min to 120 min of endurance swimming daily for 4 consecutive weeks, with a weekly increase in intensity (90, 100, 110, and 120 min at weeks 1, 2, 3, and 4, respectively). The rats in the Oil + S group were administered essential oil via inhalation during the recovery period after each swimming exercise. At the end of the 4-week experimental period, the animals were subjected to fasting for 6 h and subsequently subjected to the final endurance swim test, followed by essential oil inhalation during the recovery period and euthanasia by CO_2 asphyxiation. The livers were harvested and stored in liquid nitrogen until analysis of biochemical parameters using liver samples based on previous studies (18, 24). We found that the various antioxidant and anti-inflammatory data evaluated using liver and skeletal muscle samples revealed positive correlations after swimming training in rats. Therefore, we chose the liver as the experimental sample in this study.

Biochemical Analysis of Anti-Fatigue Indicators in the Blood

The energy metabolism markers analyzed included plasma glucose (GLU) and free fatty acids (FFAs), and the fatigue markers included lactate (LA), lactate dehydrogenase (LDH), and creatine kinase (CK). Approximately 15 mL of whole blood was collected directly from the aorta, placed in a heparinized tube, gently mixed, and centrifuged at 3500 rpm (1917 × *g*) for 15 min at 4°C. The samples were then packed in dry ice for storage and sent to a certified laboratory within 30 min for analysis.

Analysis of Antioxidation Indicators

The antioxidation indicators analyzed included markers of oxidative damage [xanthine oxidase (XO), malondialdehyde (MDA), and glutathione (GSH)], markers of antioxidant capacity [superoxide dismutase (SOD), CAT, and GSH peroxidase (GPx)], and a marker of inflammatory reactions [myeloperoxidase (MPO)]. Liver tissue from the rats was analyzed using commercially available assay kits (BioVision, Milpitas, CA, USA) to measure each of the above indicators.

Statistical Analyses

The experimental data are presented as the mean ± standard error of the mean (SEM). Data were analyzed using one-way analysis of variance, followed by Scheffe's method. A $P < 0.05$ indicated significance. SPSS software (v10.1; SPSS Inc., Chicago, IL, USA) was used for statistical analyses.

Results

Swimming Performance and Changes in Concentration

Table 1. Swimming exercise performance and changes in plasma concentration of markers associated with energy metabolism and fatigue.

Fatigue indicator	Groups			
	C	0 Oil	2w + Oil	4w + Oil
Exercise performance (s)		493.67 ± 14.25	539.83 ± 7.11*	604.67 ± 20.55**
GLU (mg/dl)	101.30 ± 11.90	101.50 ± 14.20	99.20 ± 9.40	110.80 ± 15.20
FFA (mM)	0.29 ± 0.07	0.34 ± 0.08	0.36 ± 0.07	0.39 ± 0.07
LA (mg/dl)	29.10 ± 3.00	38.40 ± 6.30*	29.20 ± 2.80	26.50 ± 3.80
CK (U/l)	1286.83 ± 53.05	1425.67 ± 45.39*	1291.00 ± 44.29	1340.50 ± 42.73
LDH (U/l)	1999.16 ± 11.58	2271.67 ± 180.83*	1998.17 ± 15.46	2044.17 ± 78.43

Groups: C, control; 0 Oil, no oil inhaled; (2w + Oil), oil inhaled for 2 weeks; 4w + Oil, oil inhaled for 4 weeks. Data for each group are shown as the mean ± standard error of the mean (SEM).
*0 Oil group was significantly different from the C, 2w + Oil, and 4w + Oil groups ($P < 0.05$).
**4w + Oil group was significantly different from the 0 Oil and 2w + Oil groups ($P < 0.05$).
CK, creatine kinase; FFA, free fatty acid; GLU, glucose; LA, lactate; LDH, lactate dehydrogenase.

of Plasma Biomarkers are Associated with Energy Metabolism and Fatigue

The weight-bearing swim-exhaustion time of the 2w + Oil and 4w + Oil groups significantly increased by 46 s and 111 s, respectively, as compared with that of the 0 Oil group ($P < 0.05$) (Table 1), whereas the plasma concentrations of the energy metabolism markers GLU and FFA did not significantly differ between the groups ($P > 0.05$). The concentrations of the fatigue markers LA, LDH, and CK were significantly higher in the 0 Oil group relative to those in the other three groups ($P < 0.05$); however, compared with that of the C group, the 2w + Oil and 4w + Oil groups showed no significant difference in these three indicators ($P > 0.05$).

Changes in Antioxidant and Anti-Inflammation Indicator Concentrations in Rat Liver Tissues

As shown in Fig. 1, the enzymatic activities of XO, MDA, SOD, CAT, GPx, and MPO in the liver tissue of the S group were significantly higher than those in the liver tissues of the other groups ($P < 0.05$). Additionally, GSH concentration was significantly lower in the S group than in the other three groups ($P < 0.05$). None of the seven indicators exhibited a statistically significant difference between the Oil + S group and the C group ($P > 0.05$).

Discussion

Anti-Fatigue Effects of Eucalyptus Oil Aromatherapy

In this study, the average weight-bearing swim time of rats after essential oil inhalation increased by an average of 46 s and 111 s (2w + Oil and 4w + Oil, respectively), indicating a clear improvement in exercise performance (Table 1). During high-intensity endurance exercise, liver glycogen is catabolized, releasing GLU into the blood, which delivers GLU to the muscles to provide energy. Fatigue can result if the blood GLU content is low (22). Lipolysis results in the metabolism of lipids into FFAs, thereby increasing their utilization as energy sources (15). Therefore, the GLU and FFA content in the blood can serve as indicators of high-intensity endurance-training ability. The results of this study showed that the GLU and FFA values in each group after swimming did not significantly differ. Therefore, it is likely that the anti-fatigue effect of essential oils on exercise performance in rats is not directly related to carbohydrate or lipid energy metabolism.

LA represents one of the products resulting from energy metabolism during muscle activity, and its accumulation is associated with fatigue. After high-intensity endurance exercise, muscle cells undergo accelerated glycolysis due to the severe lack of oxygen, leading to lactic acid accumulation, increased hydrogen ion concentration, lower pH, and decreased phosphofructokinase activity. Furthermore, these processes affect Ca^{2+} release, thereby decreasing the ability of muscle fibers to contract and accelerating fatigue. In the present study, LA significantly decreased in the essential oil inhalation groups (2w + Oil and 4w + Oil). This result might be related to the finding that 1,8-cineole, the primary component of eucalyptus

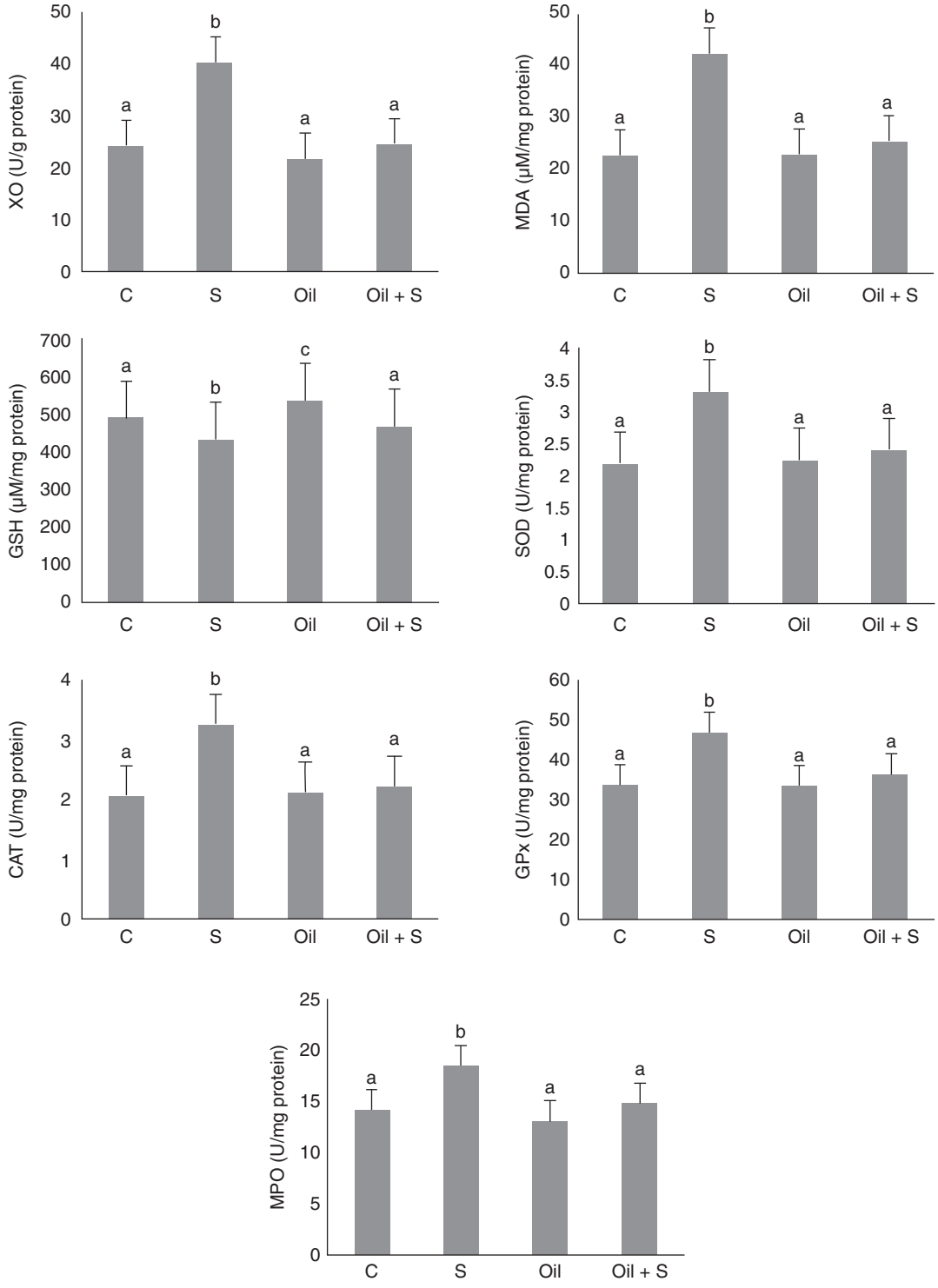

Fig. 1. Changes in concentrations of antioxidant and anti-inflammation indicators in rat liver tissues. Values [representing the mean ± SEM (n = 8) for all test groups] in each column not sharing the same alphabetic letter are significantly different ($P < 0.05$). CAT, catalase; GPx, GSH peroxidase; GSH, glutathione; MDA, malondialdehyde; MPO, myeloperoxidase; SOD, superoxide dismutase; XO, xanthine oxidase.

oil, accelerates blood flow (7).

In particular, results similar to those obtained in the present study were observed in an experiment with Shin-I essential oil, which also contains 1,8-cineole as its primary component (7). Rats that inhaled Shin-I essential oil showed a 28% increase

in treadmill exercise performance along with significant decreases in LA concentration in the blood (7), revealing that 1,8-cineole exerted significant pharmacological effects by accelerating blood flow. This occurs through the rapid penetration of the nasal mucosa and activation of the olfactory center, which further affects neurotransmitter secretions (31, 34). This activity can cause vascular smooth muscle relaxation through parasympathetic nerve-dependent induction of hypotensive effects and vasodilation (5, 23). Additionally, local application of 1,8-cineole promotes blood circulation, which results in skin hyperemia (5). Therefore, the results of the present study suggest that 1,8-cineole in eucalyptus oil causes vasodilation, increases blood flow, and promotes blood circulation, thereby increasing the clearance rate of LA, as well as exercise performance.

The primary function of CK in blood plasma is to support the production of energy needed during exercise, whereas LDH catalyzes the conversion of pyruvate into LA. In addition to affecting aerobic and anaerobic metabolism during exercise, an abundance of LDH and CK is also an important indicator of tissue damage. Decreased muscle strength caused by slight tissue damage is closely related to exercise-induced fatigue. High-intensity weight-bearing exercise can significantly increase CK and LDH levels, which in turn decrease exercise performance (11). In the present study, the activities of blood plasma CK and LDH in the 0 Oil group were significantly higher than those in the other three groups. Conversely, the two essential oil inhalation groups (2w + Oil and 4w + Oil) did not exhibit a significant difference in activity from that observed in the C group. This showed that eucalyptus oil decreased the extent of skeletal muscle damage after swimming exercise, which might have been related to changes in oxygen free radicals during such exercise.

Increased oxygen uptake during high-intensity endurance exercise promotes free radical production, excessive amounts of which can cause muscle fatigue (3). In a study of the effects of strenuous exercise on the soleus muscle of rats, the free radical signal in the muscle was elevated 2- to 3-fold after exercise, with the increase in free radicals found to be closely related to damaged cell-membrane integrity (9). Therefore, the anti-fatigue effects of eucalyptus oil might also be related to the antioxidant effects associated with the involvement of its active components in decreasing LDH and CK concentrations. These actions reduce the extent of skeletal muscle damage after swimming exercise, which in turn alleviates muscle fatigue.

Antioxidant and Anti-Inflammatory Effects of Eucalyptus Oil Aromatherapy

Reduction of oxidative damage from swimming exercise. In addition to the mitochondrial electron-transport chain, the primary source of free radicals in cells during exercise is XO activity in the muscles, which increases notably under these conditions. The conversion of adenosine triphosphate (ATP) into adenosine diphosphate and adenosine monophosphate (AMP) fulfills the energy requirements for muscle contraction. Therefore, a lack of oxygen prevents the production of ATP by oxidative phosphorylation, and AMP undergoes further degradation to hypoxanthine. Increased XO activity catalyzes the oxidation of accumulated hypoxanthine into xanthine and large amounts of uric acid and promotes oxidative damage by $O_2^{\cdot-}$ production (40). Additionally, increased XO activity due to high-intensity anaerobic exercise leads to oxidative damage to other tissues and organs *via* the blood (41).

In the present study, 4 weeks of endurance-swimming exercise in rats increased XO activity and lipid peroxide production in liver tissues. This was confirmed in the group of rats that underwent endurance swimming (S), whereas liver-tissue XO activity was significantly higher than that in the control group (C; $P < 0.05$). MDA exhibited similar results, because liver-tissue MDA content was significantly elevated in the S group as compared with that in the C group ($P < 0.05$). However, in the Oil + S group, XO activity and MDA content in the liver tissue were not significantly elevated as compared with that of the C group ($P > 0.05$).

GSH in its reduced form is an important protective factor that plays a critical antioxidant role in the body. Moreover, it represents an important biological indicator of the prevention of oxidative damage and fatigue caused by exercise (32). Studies show that high-intensity exercise increases oxidized GSH (GSSG) and decreases both total GSH and the GSH:GSSG ratio in cardiac and skeletal muscles, as well as in the liver (26). The acute oxidation reactions that occur during high-intensity endurance training increase intracellular GSH reductase content; however, under these conditions, energy consumption acutely increases and competes with the GSSG-reduction pathway for NADPH. Ultimately, GSSG production exceeds reduction capacity, resulting in decreased GSH content in various tissues (26).

High-intensity endurance training leads to the production of large amounts of free radicals, which damage numerous enzymes and membrane structures. This in turn affects GSH synthesis because of insufficient precursor availability or damage to its synthesis enzymes. Simultaneously, during high-intensity endurance training, glycogen is exhausted,

ATP synthesis is reduced, and NADPH produced by the pentose phosphate pathway is reduced. These actions also affect GSH synthesis and lead to decreased GSH concentrations in the liver. The relatively insufficient release of GSH combined with the increased degradation of GSH in tissues during exercise ultimately results in decreased GSH content in the body.

In the present study, GSH content in the liver significantly decreased in rats in the endurance swimming (S) group after 4 weeks of endurance-swimming exercise as compared with that in the C group ($P < 0.05$). By contrast, the Oil + S group did not show a significant difference from the C group. Additionally, GSH content in the liver tissue of rats in the C group was significantly lower than that in the essential oil group (Oil; $P < 0.05$). These results showed that the natural antioxidants contained in eucalyptus oil effectively scavenged free radicals, thereby reducing GSH consumption. In the present study, the GSH content in the liver tissue of rats that inhaled essential oil remained at a relatively high level after 4 weeks of exercise.

Increased antioxidant Capacity after endurance-swimming training. SOD is an endogenous antioxidant enzyme that serves as a first-line defense against free radical production. During the initial reactions of free radical formation, it transforms $O_2^{\cdot -}$ into H_2O_2 and scavenges ROS in combination with GPx, CAT, and other antioxidant enzymes. After endurance-swimming training in rats, SOD activity immediately increases in skeletal muscle, cardiac muscle, and the liver (19, 38). This result is consistent with that observed in the endurance-swimming (S) group in the present study. The increase in SOD activity is primarily due to the increased production of $O_2^{\cdot -}$ (39); however, a previous study also showed that high-intensity exercise in rats significantly decreased SOD activity in red blood cells (16). Therefore, SOD activity is regulated differently in different tissues. Moreover, liver-tissue SOD activity in the essential Oil + S group did not differ significantly from that in the C group, suggesting that eucalyptus oil had a protective effect against oxidative stress caused by endurance-swimming training.

A previous study showed that CAT activity increases in the heart, skeletal muscle, and liver after endurance training (1), which is consistent with the liver-tissue results found in the present study. Furthermore, CAT activity significantly increased in the endurance-swimming (S) group as compared with that in the C group ($P < 0.05$). Similar to SOD activity, CAT activity in liver tissues from the Oil + S group showed no significant difference from that in the C group after exercise ($P > 0.05$). This indicated that CAT activity did not significantly increase after endurance-swimming training when the rats were pretreated with eucalyptus oil *via* inhalation. Furthermore, the protective effect of the active components of eucalyptus oil likely reduced the damage caused by H_2O_2 accumulation in cells.

GPx exists in the cytoplasm and mitochondria and protects cellular proteins, lipids, and nucleic acids from free radical-induced damage (29). The GSH:GSSG ratio in red blood cells decreases significantly after endurance training in rats (21). Additionally, a previous study indicated that GPx activity in the blood significantly increases in rats after endurance training, and that GSH activity either decreased significantly or showed no significant change (33). For other tissues, such as cardiac muscle, skeletal muscle, and the liver, contradictory results were obtained regarding GPx activity (17). These inconsistent experimental results might be due to the different regulatory mechanisms associated with ROS production in various tissues that lead to differences in antioxidant enzyme activity. Furthermore, rats inherently synthesize vitamin C and GSH, which can also affect antioxidant enzyme activity, and levels of ROS production in different tissues can be affected by the experimental subject, exercise intensity, exercise duration, and other factors (2). The results of the present study showed that GPx activity in the liver tissue of the S group was significantly elevated after endurance-swimming training, and that there was no significant difference between the Oil + S and C groups ($P > 0.05$). The increased GPx activity in the S group suggested that one of the functions of the liver in compensating for oxidative stress caused by endurance-swimming training is to scavenge ROS produced after exercise. In the Oil + S group, essential oil inhalation did not significantly change antioxidant enzyme activity before or after exercise, indicating that eucalyptus oil protected against oxidative stress caused by endurance-swimming training.

Reduction of inflammatory reactions following endurance-swimming training. Strenuous exercise produces acute inflammation of muscle tissue. Detection of foreign-matter invasion by the immune system also involves the initiation of oxidative stress (10). Inflammatory reactions initially activate neutrophils, which increase during and after strenuous exercise, and increases NADPH oxidase activity, leading to the production of $O_2^{\cdot -}$ and H_2O_2. MPO is released by neutrophils and uses H_2O_2 to produce the even stronger oxidizing agent, hypochlorous acid. As a result, inflammatory reactions induce oxidative stress and produce large amounts of ROS (37). Therefore, MPO is a biological indicator of inflammation caused by neutrophil invasion of tissues. In previous animal studies, rats showed significant increases in

MPO activity in the heart, liver, and skeletal muscle after exhaustive exercise (4). This result was confirmed in the present study. Conversely, liver-tissue MPO activity in the Oil + S group showed no significant difference relative to that in the C group, indicating that eucalyptus oil inhalation effectively inhibited inflammatory reactions involving neutrophils.

Eucalyptus oil aromatherapy increased the swimming performance of rats. The anti-fatigue effects of the essential oil are primarily due to the degree of skeletal muscle damage, as indicated by LA clearance and decreased LDH and CK activity after swimming exercise. Eucalyptus oil aromatherapy reduced XO, MDA, and MPO production in the liver tissue, as byproducts of endurance-swimming training, and maintained GSH, SOD, CAT, and GPx stability, thereby alleviating oxidative damage and inflammation in tissues. Eucalyptus oil aromatherapy exhibited good anti-fatigue, antioxidant, and anti-inflammatory effects in rats after swimming exercise. However, further studies are required to validate whether eucalyptus oil can cause similar effects in humans when used as adjuvant therapy. The findings of our study lay the foundation for future studies in sports science and athletic training, as well as for studies regarding the application of eucalyptus oil aromatherapy during training to improve athletic performance.

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Conflict of Interests

The authors declare that there are no conflicts of interests.

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