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The Comparison of Beet Juice and Ibuprofen Consumption on Muscle Damage after a Session of Resistance Exercise in Young Untrained Women

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ABSTRACT

This study aimed to compare the consumption of two combinations of beetroot juice and ibuprofen before exercise on muscle damage indicators after a session of isotonic resistance exercise. The research samples consisted of 24 young women (20-34 years old) who were randomly divided into two groups of beetroot juice (n=12) and ibuprofen (n=12). The beet juice group received 4.5ml of natural beet juice per kilogram of body weight about 150 minutes before the activity, and the ibuprofen group received 30mg of the drug 24 hours before the activity in three meals. Both groups performed resistance activities, including six movements of the upper body and lower body in three times of 15 to 20 repetitions with 60 to 70% of the maximum strength. In the end, the levels of creatine kinase and blood myoglobin were evaluated in two stages. The data were analyzed using the independent t-test at an error level of p ≥0.05. Although the serum creatine kinase and myoglobin levels in both groups showed a significant increase compared to pre-activity (p=0.0001), no difference was observed between the groups (p=0.094 and p=0.211). In addition, the concentration of this variable in the ibuprofen consumption group had a slight change, while the increase in plasma interleukin-6 concentration in the beetroot juice consumption group had a significant increase compared to before the activity (p=0.001). The results showed that ibuprofen reduces the inflammation caused by these exercises to some extent.

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Introduction

Resistance training is one of the suitable training methods for women, which helps them improve their muscle tone and muscle readiness in addition to increasing their energy consumption (1). Intense or unusual sports activities can cause muscle damage, which is called exercise-induced muscle damage (EIMD). An initial response characterizes muscle damage caused by exercise as a result of mechanical stress that occurs during exercise and the response of secondary inflammatory indicators (2). Mechanical force, especially in extrinsic activities (muscle contraction with increased muscle tension), leads to the initial response. Precisely, overstretching and disruption of sarcomeres, followed by increased Ca^{2+} influx into muscle cells, leads to passive muscle tension and myofibrillar dysfunction (3). The responses subsequently trigger secondary inflammatory reactions, including the production of reactive oxygen species (ROS) and cytokines by promoting the activation of transcription factors (e.g., NF-κB[1], MAPK[2] and (Nrf2[3]), stimulate. In addition, reactive oxygen species (ROS) and cytokines can be released from phagocytic neutrophils and macrophages (4).

Exercise-induced muscle injury (EIMD) appears to be characterized by several symptoms such as loss of muscle function, e.g., reduced strength and decreased range of motion, DOMS, and increased leakage of muscle proteins such as creatine kinase, myoglobin, and aspartate transaminase,

1. Nuclear Factor-kappa B
2. Mitogen-Activated protein kinase
3. Nuclear Factor erythroid 2-related factor 2

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enter the blood circulation (5), which can reduce athletic performance. Therefore, reducing these symptoms is also important to optimize sports performance and physical fitness. Several nutritional strategies have been proposed to restore muscle function, relieve delayed onset muscle soreness (DOMS), and reduce post-exercise inflammation. According to the International Olympic Committee (IOC) Agreement on Nutritional Supplements, several nutritional supplements are recommended for high-performance athletes, including creatine monohydrate, beta-hydroxy beta-methyl butyrate, omega-3 fatty acids, vitamin D, gelatin, vitamin C/Collagen, as well as anti-inflammatory supplements, such as curcumin and sour cherry juice, for improving exercise capacity, recovery, muscle soreness, and injury management (6). Among these supplements, anti-inflammatory supplements have been suggested to reduce muscle injury and soreness (7). Reduction of delayed-onset muscle soreness (DOMS) may be essential in sports activities, as such pain may interfere with subsequent periods of exercise (8). Furthermore, both inflammatory responses, reactive oxygen species, and free radicals produced during and after exercise have been suggested to be involved in DOMS (13). Therefore, nutrition-based interventions that target post-exercise inflammation and/or oxidative stress responses (imbalance between free radicals and antioxidants in the body) have received much attention.

Researchers are focusing on replacing chemical drugs effective in reducing pain and inflammation with low-risk natural compounds because of the numerous side effects of chemical drugs. Researchers have focused on easy access. For example, in the pharmacological research conducted on delayed muscle contusion, natural and non-synthetic antioxidants and other natural antioxidants such as vitamins E and C have been used (9). A review of supplements used for reducing EIMD and relieving delayed-onset muscle soreness (DOMS) in humans is presented, focusing on those that have been cited in IOC statements as anti-inflammatory and/or antioxidant. The ingredients include curcumin, sour cherry juice, beet juice, and isothiocyanate, which are all anti-inflammatory or antioxidant. In addition, in this study, among the foods rich in nitrates, beetroot juice (aside from nitrates, beetroot is a rich source of powerful antioxidants like vitamin C, carotenoids, phenolic acids, and flavonoids. Besides being rich in sugar and phenolic compounds, ascorbic acid is highly soluble in NO3 and is consumed because it contains betacyanin and polyphenols in high amounts. Among the anti-inflammatory drugs, ibuprofen (for Accelerating recovery processes and preventing muscle soreness, the effects of NO3-rich sources were first observed in metabolic adaptations after endurance exercise), and other techniques (such as cryotherapy and non-steroidal anti-inflammatory drugs (NSAIDs) (ibuprofen) during training and competition) were discussed.

Although non-steroidal anti-inflammatory drugs (NSAIDs) are often used to reduce exercise-induced muscle soreness and accelerate post-exercise recovery, there is no support for such an effect in the history of athletes. Therefore, the present study seeks to answer the question of the differences between the consumption of beetroot juice as a natural combination and the consumption of non-steroidal anti-inflammatory drugs before a resistance activity on muscle damage and inflammation indicators in untrained women.

Materials & Methods

Subjects

The statistical samples of this study consisted of 24 young women without training who were in the age range of 20 to 34 and who did not have any injuries or chronic inflammation, food and drug allergies, coagulation disorders, diabetes, immune system disorders, and other problems, including digestive, cardiovascular, and respiratory. The samples voluntarily declared their readiness to participate in the present study and completed the personal consent form to participate in the research after being fully aware of how the research was conducted and provided it to the researcher. After preliminary investigations, they were selected and randomly divided into two groups of 12 people, which included the group using beetroot juice (BJ) and the group using ibuprofen (PL).

Procedure

Volunteers were invited to participate in the research after the invitation was issued to implement the present study. Preliminary investigations were conducted to select the samples, and 24 subjects were selected from among the volunteers. A consent form to

participate in the survey was completed after a meeting explaining how the research would be conducted and possible risks. After that, the subjects were taught the initial evaluations and resistance training movements, including six common movements in resistance training programs (1). In the following, initial evaluations about the maximum strength of the subjects (estimation of one repetition maximum) were taken and recorded from the subjects. For the purpose of controlling the nutritional factor, the subjects were instructed to consume three units of fruits and vegetables in their diet one day before the research. Also, the night before the implementation, they ceased consuming fruits and vegetables and any anti-inflammatory medicines.

A total of 24 hours before the resistance exercise, the samples of the two groups received the supplements after 90 minutes. After the last meal of receiving the supplements, blood was taken from the samples. Blood sampling was performed from the brachial vein of the samples 15 minutes before the performance of resistance exercise, and then they performed a session of resistance activity. A second blood sampling was conducted 15 minutes after the end of the activity, and a second blood sampling was performed 24 hours after the activity to evaluate the dependent variables.

Preparation of Beetroot Juice and How to Prepare Supplements

Research has shown that beets were first cleaned three to four times with fresh water to prepare beetroot juice. Then, the beets were completely peeled and cut, and their juice was extracted using a hydraulic juicer and filtered with a double-layer strainer. This liquid was pasteurized in a steel container at 96°C for 720 seconds and then transferred to cold water with an approximate temperature of 2°C for 2 to 3 minutes. Then, it was stored in a sterile glass container at 4°C in the refrigerator until use, and on the research day, 4.5 grams per kilogram of weight was given to the samples of the beet juice consumption group (10). The subjects took 2800mg of ibuprofen from one hour before the outward contractions to 47 hours after the end. This medicine was given to the subjects in seven doses of 400mg (11).

Statistical Methods

In the statistical analysis section, the Shapiro-Wilk test was used to determine the normality of the data distribution. After assessing the normality of the data, the independent t-test was used to compare the changes in the dependent variables in the two groups. The error level considered for analyzing the results was also considered as $\alpha \leq 0.05$.

Research Findings

The first part of the research findings is related to the general characteristics of the samples, which are listed in Table 1. As shown in Table 1, the average and standard deviation related to the three indicators of age, height, weight, and body mass index of subjects of the two groups are:

The results show that all three blood variables measured in the groups reveal a significant difference 2 hours after the activity and 24 hours after the activity. In addition, the value of this difference in interleukin six variable, 2 hours after the activity and 24 hours after the activity, indicates a significant difference between the groups.

Table 1. Individual characteristics of subjects in research groups

Individual characteristics	ibuprofen group variable value	Beet juice group, variable value
Number of subjects	12	12
age (years)	26.21±4.19	25.67±3.57
weight (kg)	59.38±5.71	61.46±6.93
height (cm)	164.08±3.13	165.46±5.36
BMI (K/m^2)	22.01±2.71	22.28±3.50

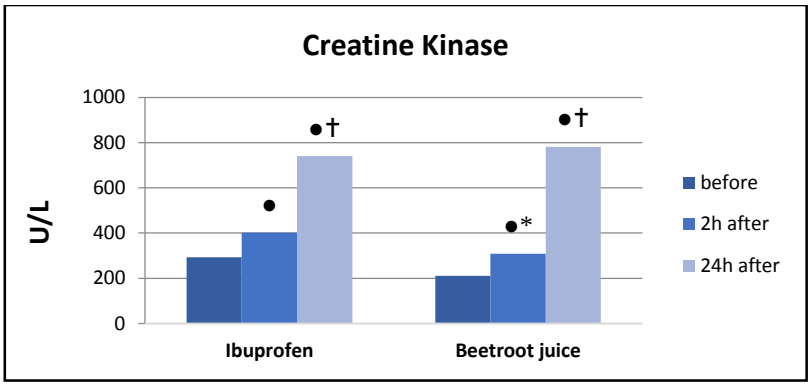

Figure 1. The average values of serum creatine kinase concentration before, 2 hours after and 24 hours after the activity in the research groups
(*) significant difference with before the activity at level of p≤0.05.
(●) Significant difference with 2 hours after activity at level of p≤0.05.
(†) significant difference with the beet juice group at the level of p≤0.05.

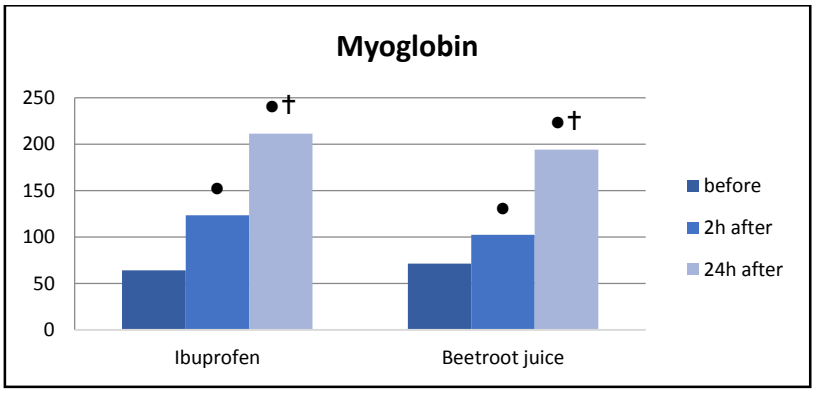

Figure 2. The average values of serum Myoglobin concentration before, 2 hours after and 24 hours after the activity in the research groups
(*) significant difference with before the activity at level of p≤0.05.
(●) Significant difference with 2 hours after activity at level of p≤0.05.
(†) significant difference with the beet juice group at the level of p≤0.05.

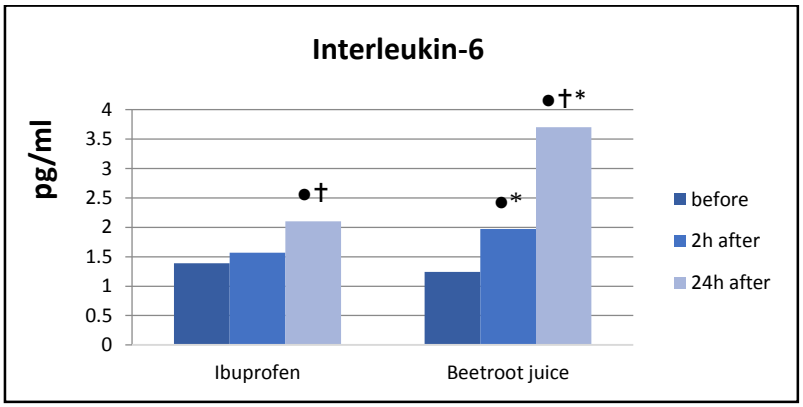

Figure 3. The average values of serum Interleukin-6 concentration before, 2 hours after and 24 hours after the activity in the research groups
(*) significant difference with before the activity at level of p≤0.05.
(●) Significant difference with 2 hours after activity at level of p≤0.05.
(†) significant difference with the beet juice group at the level of p≤0.05.

Discussion

There was no significant difference between beetroot juice and ibuprofen in inhibiting muscle damage and its indicators, namely myoglobin and serum creatine kinase, 2 hours after the activity and 24 hours later. This study was planned based on assuming the effectiveness of beetroot juice consumption in reducing pain and bruising caused by a resistance activity session. These results were consistent with those of Jones et al., who showed in a review study that the consumption of beetroot juice reduces some indicators of muscle damage after outdoor activities (12). On the other hand, these results were in line with those of Tanabe et al. (2021). The mechanisms involved in the occurrence of these results have been attributed to the antioxidant effects of anthocyanin (1). Studies on animals, humans, and laboratory studies have reported powerful vascular protective antioxidant and anti-inflammatory effects from beetroot. The antioxidant capacity of beetroot juice is much higher than that of more famous vegetables such as tomato, carrot, orange, and pineapple juice. Beetroot contains a solid and powerful antioxidant called anthocyanin, which is located in its purple-red skin and pith. For this reason, one of the default bases of this study was the antioxidant effect of beetroot juice in inhibiting inflammation and muscle damage caused by a session of resistance activity. On the other hand, beetroot is used as a traditional medicine to treat constipation and fever. Moreover, beetroot juice is used as a tonic to strengthen sexual powers, and it is rich in nitrates. However, the current study showed that consuming beetroot juice probably does not reduce muscle tissue damage, but it may reduce the resulting inflammation to some extent.

Nitric oxide is an essential physiological messenger molecule that can alter skeletal muscle function through its role in regulating blood flow, contractility, glucose and calcium homeostasis, respiration, and mitochondrial biogenesis (14). The amino acid L-arginine is produced in a reaction catalyzed by nitric oxide synthase, and nitrite and nitrate are inert by-products produced in this process (15). However, it is now known that these metabolites can be converted to nitric oxide under certain physiological conditions, which is biologically active and can be recycled (16). The enzyme nitric oxide synthase can disrupt nitric oxide synthesis in conditions with low oxygen availability, which may make the reduction of nitrate to nitrite and subsequent conversion of nitrite to nitric oxide an essential tool for increasing nitric oxide production. In addition to the production of nitric oxide from L-arginine through the reaction catalyzed by nitric oxide synthase, the tissue concentration of nitrate and nitrite can be increased by using diet. In the Western diet, vegetables account for about 60-80% of the daily intake of nitrates. Green leafy vegetables such as lettuce, spinach, and beets are rich in nitrates (17). Inorganic nitrate, eaten, is quickly absorbed in the intestine and enters the general circulation. Plasma nitrate concentration reaches its maximum value about 60 minutes after consumption. However, about 60% of nitrates in general circulation are excreted through urine (16). However, about 25% enters the saliva, and for this reason, the saliva has a high concentration. In the mouth, various anaerobic bacteria on the surface of the tongue reduce nitrate to nitrite (5). After digestion, this nitrite is reduced to nitric oxide, and other reactive nitrogen intermediates are reduced in the stomach's acidic environment. However, some of the nitrite is absorbed to increase its plasma concentration, and about 23 hours after consuming, the nitrate concentration reaches its peak value (18). Therefore, dietary nitrate supplementation is a practical way to increase the concentration of circulating nitrite and thus increase the availability of nitric oxide. Increased access to nitric oxide has been observed after consuming sodium nitrate (11) and potassium nitrate (19), as well as beet juice rich in nitrates (20). Although the subjects of the beetroot juice group may have better blood circulation in the muscles involved in the activity due to receiving higher nitrates, a definitive statement cannot be made since the index of measuring the blood circulation of the muscles during the activity was not measured in the present study. The results of the present study also showed that the concentration of interleukin-6 as an inflammatory cytokine index after exercise was lower in the group consuming ibuprofen than in the group consuming beet juice. Therefore, ibuprofen, as a non-steroidal anti-inflammatory drug, has been able to partially reduce the inflammation caused by tissue damage after exercise. However, beetroot juice supplementation has not had much effect on this

inflammation. Ibuprofen is a non-steroidal anti-inflammatory drug, and it is believed that it works by inhibiting cyclooxygenase (Cox), thus inhibiting the production of prostaglandins. There are at least two different cyclooxygenases (1-Cox-2. Cox). Ibuprofen inhibits both, and its antipyretic and anti-inflammatory properties seem to be related to the inhibition of 2-Cox (6). However, Fraga et al. (2020) showed that ibuprofen reduced inflammation up to 24 hours after the activity, but after this time, inflammation increased in subjects taking ibuprofen. This position indicates that the use of ibuprofen only delays the inflammation and cannot reduce it in general. Although receiving beetroot juice and ibuprofen before performing resistance sports activities can probably lead to inhibition of indicators related to muscle damage, there is a difference between the effect of these two supplements on indicators such as creatine kinase and myoglobin. There was no serum, but the use of ibuprofen was able to partially inhibit the inflammation in the active muscle tissue up to 24 hours after the activity, and it is not clear whether this effect is maintained after 48 and 72 hours after the activity or not. Future studies can better evaluate this issue. Further studies should assess and monitor inflammation variables up to 72 hours after the end of the activity in future studies.

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