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The Effects of Consuming 6 Weeks of Beetroot Juice (*Beta vulgaris* L.) on Hematological Parameters in Female Soccer Players

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Abstract

Background: The present study aimed to investigate the effects of consuming 6 weeks of beetroot juice (*Beta vulgaris* L.) on hematological parameters in female soccer players.
Methods: Twenty female soccer players were selected randomly and assigned into two groups: Experimental (beetroot juice, n = 10) and control (placebo, n = 10). Subjects trained for six weeks (three 90-min sessions per week) by consuming 200 mL of juice 2 hours prior to training. Blood samples were collected and investigated (Hb, Hct, RBC, Iron, MCV, Ferritin and TIBC) in pre- and posttest. Paired Sample *t*-test and Independent Sample *t*-test were used for comparision within and between groups, respectively. Statistical significance was $P \leq 0.05$.
Results: Results showed that the experimental group had significant increases in the Hb, Hct, RBC, iron, and ferritin levels ($P < 0.05$). No significant difference was detected in MCV levels ($t = -1.10$, $P = 0.29$) and there was a significant decrease in the TIBC ($t = 4.99$, $P = 0.001$). In the control group, there were significant differences in (Hct, iron, ferritin and TIBC) ($P < 0.05$). In addition, there were significant differences in (Hb, Hct, RBC, iron, ferritin and TIBC) levels between experimental and control subjects ($P < 0.05$), but there was no significant difference in MCV ($t = 1.11$, $P = 0.28$).
Conclusions: Since beetroot juice consumption has significant effects on the levels of some hematological parameters in female soccer players, it can be used to prevent and improve anemia among these athletes.

Keywords: Anemia, Female Adolescents, Beetroot Juice, Soccer

1. Background

Soccer is one of the most favorite sports played by men and women with different levels of fitness. Soccer players cover a distance of 10 - 12 km. Cardiorespiratory endurance is one of the main elements of physical fitness (1). Aerobic fitness shows an integrated action with cooperation from the lungs, heart, blood and active muscles. More recently, it is believed that hematological parameters may also play a vital role in anticipating optimal performance in soccer players. Hematological parameters are affected by several factors include training, age, sex, ethnicity, nutrition, and altitude (2). Anemia is a common disorder among adolescent women regardless of levels of physical activity and defined as a decreased concentration of blood hemoglobin. It has a variety of converging contributing factors including nutritional, genetic and infectious disease factors (3). Although many causes of anemia have been known, it is believed that iron deficiency is an important cause. Iron is an important element that makes up the heme molecules of hemoglobin in red blood cells, allowing for oxygen transportation and utilization (4). During anemia, defined as a deficiency of red blood cells, a decrease of hemoglobin impairs oxygen delivery to the tissues, predisposing to performance decrements despite a compensatory increase of cardiac output (5). Yet, elite athletes, especially female soccer players have a higher risk of iron depletion during intensive training due to several possible mechanisms (i.e. intravascular/foot-strike hemolysis (4, 6), ischemia to the viscera (7) or frequent nonsteroidal anti-inflammatory drug use (8) which may promote gastric lesions (7), loss of iron through urine caused by renal ischemia (6), menstrual cycle, etc. Evidence propose that depletion of iron stores are associated with decreased aerobic performance. Detection of depleted iron stores in early stages seems to be a successful intervention for preventing iron deficiency and anemia (9). Adolescent

female soccer players with anemia have a diminished athletic performance. Nutritional supplements and organic materials are best for eliminating iron depletion and anemia in female soccer players. Beetroot (*Beta vulgaris* L.) is a main source of iron, nitrate, sodium, potassium, and betalain among vegetables (10). Lowering blood pressure by dilating the blood vessels and relaxing smooth muscles, treating anemia by increasing the blood count and improving blood circulation and the oxygen-carrying capacity of erythrocytes (red blood cells), preventing birth defects by increasing folate and folic acid, preventing hypertension and strokes, cleansing the intestines, reducing kidney stones, improving rheumatoid arthritis, gout sand improving menstrual problems are the benefits of beetroot juice intake (11). According to Bakhru, author of "*Foods That Heal*", beetroot juice is particularly beneficial for treating anemia in children and teenagers. Easton Patrick (2011) believes that consuming beetroot juice or cooked beet in salads is highly beneficial in treating anemia. Nirman Walker (2010) claimed that beetroot builds red corpuscles with betalain and adds tone to blood by increasing hemoglobin levels. The cost is low as compared to other iron-rich vegetables and it is easy to store (12). Thus, the aim of this research was to evaluate the effects of 6 weeks beetroot juice (*Beta vulgaris* L.) consumption on hematological parameters in female soccer players.

2. Methods

The study was conducted at Razi University, Kermanshah, Iran. The research design was a kind of semi-experimental design. The study protocol received ethical approval from the Faculty of Sports Sciences Ethics Committee at Razi University.

2.1. Subjects

Written permission was obtained from the subjects after explaining the purpose of the study. None had any known food allergies, thalassemia (minor, major), blood cancer, unknown blood diseases, previous history of renal, gastrointestinal or cardiovascular complications or any other contraindications to the study procedures. Subjects were asked not to use supplements, coffee, energy drinks (13) and drugs that affected the blood profile such as anticoagulant drugs like heparin, antiplatelet drugs like aspirin, thrombolytic drugs like streptokinase, drugs for treating anemia like iron and folic acid, and drugs for treating bleeding like thrombin. Subjects had the same diet and ate food provided by the central canteen at Razi University.

The sample size selected for this study consisted of 20 adolescent female soccer players' 23.13 $\pm$ 0.77 years old, BMI: 23.05 $\pm$ 1.01 kg/m^2, playing in the Razi University soccer team. Subjects were divided into two groups randomly (experimental and control: n = 10). In the first session, blood samples were collected in fasting mode. Beetroot juice and placebo (water, carmoisine food coloring and stevia = 200 mL) were given to subjects 2 hours before they started their training (14).

2.2. Background Variables

Background variables used in the questionnaire to collect data from the female soccer players included their age, type of family, family income per month, number of children in the family, type of diet, meals per day, and menstrual cycle; they were also asked if their diet contained more vegetables, if they had suffered from anemia before, and if yes, were they taking any treatment.

2.3. Anthropometric Measurements

Height, weight and BMI were obtained before the test. Height was measured by using semiautomatic height measurement equipment (HD, STDK, Tokyo, Japan); weight and body compositions were measured by a bioelectrical impedance analysis body composition analyzer (Inbody220, Biospace, Seoul, Korea) (Table 1).

2.4. Exercise Training Method

Training sessions were held three times a week for 90 min each; following general and special warm-ups (dribbling, shooting, etc.) the participants did their preparation programs before soccer competitions between universities.

2.5. Blood Collection and Analysis Method

Before and after treatment, blood (10 mL) was collected from the antecubital vein with a 22-gauge needle and placed in test tubes containing an anticoagulant substance. This was taken to the laboratory to measure the hematological parameters (Hb, Hct, RBC, Iron, MCV, Ferritin and TIBC). Test tubes were utilized for a complete

Table 1. Anthropometric Measurements (Weight, Height, BMI)[a]

Variables	Experimental Group	Control Group
Weight (kg)	58.56 $\pm$ 4.88	61.18 $\pm$ 2.48
Height (cm)	161.05 $\pm$ 5.10	162.60 $\pm$ 1.89
BMI (kg/m^2)	22.57 $\pm$ 1.54	23.13 $\pm$ 0.58

[a]Values are expressed as mean $\pm$ SD.

blood count with the UDI hemolysis machine. Blood separation was performed by centrifugation at 3000 rpm for 15 min. The levels of Hb, Hct, RBC, iron, MCV, ferritin and TIBC in pretest and posttest were compared.

2.6. Statistical Analysis

Data was analyzed with the SPSS software (version 22). The Paired Sample *t*-test was used for comparison within groups and Independent Sample *t*-test was used for comparison between groups. Statistical significance (P) was set at 0.05.

3. Results

For the type of family, 83.3% of experimental group and 80% of control group were from rich families. For family monthly incomes, 36.67% in the experimental group and 46.67% in the control group had incomes of two million tomans. For the type of diet, 90% in the experimental group and 90% in the control group were non-vegetarians. For a history of anemia, 100% of the girls in the experimental group and 100% in the control group had no history of anemia. For menstrual cycles, all the subjects in both the experimental and control groups had regular menstrual cycles.

3.1. Changes in Hematological Parameters

Changes in hematological parameters (Hb, Hct, RBC, iron, MCV, ferritin and TIBC) before and after treatment in the experimental and control groups are shown in Table 2.

Table 2 shows the results of laboratory investigations for Hb, Hct, RBC, iron and MCV from blood samples of the subjects before and after 6 weeks beetroot juice consumption during soccer training. After the administration of 6 weeks beetroot juice consumption, significant improvements were observed in (Hb, Hct, RBC, iron and ferritin) levels of subjects ($P < 0.05$), whereas there was no significant improvements in (MCV, TIBC) levels ($P > 0.05$). In the control group, there were significant differences in (Hct, iron, ferritin, TIBC) levels ($P < 0.05$), but there were no significant differences in (Hb, RBC, MCV) ($P > 0.05$).

3.2. Changes in the Hematological Parameters (Hb, Hct, RBC, Iron, MCV, Ferritin and TIBC) Between Experimental and Control Groups

Table 3 shows comparisons between mean and standard deviations made for experimental and control subjects using the Student *t*-test. Results show that there were significant differences in Hb, Hct, RBC, iron, ferritin and TIBC levels between experimental and control subjects ($P < 0.05$), but there were no significant differences in MCV between experimental and control subjects ($t = 1.11$, $P > 0.05$).

4. Discussion

In our study, hemoglobin levels significantly increased in the experimental group ($P < 0.05$), indicating the positive effects of beetroot juice on Hb levels. This finding is consistent with the studies conducted by Priya et al. (12), Nora (15) and Patel et al. (16), whereas the control group showed no significant change in Hb levels ($P > 0.05$). In this study, the experimental group had a significant increase in Hct levels as compared with the pretreatment control group. It showed us that there were great improvements in Hct levels ($P < 0.05$) after the administration of beetroot juice. Whereas, in the control group, there were significant differences in the Hct levels of adolescent female soccer players ($P < 0.05$). There was a slight decrease in the Hct levels of the control group which was related to the fact that athletes often have decreased levels of Hct called sports anemia (5). RBC levels increased significantly in the experimental group in relation to pretest levels as compared to the control group. It proved that after beetroot juice intake, there were significant improvements in RBC levels ($P < 0.05$). Whereas, in the control group, there were no significant improvements in RBC levels ($P > 0.05$). In the experimental group, there were significant improvements in the iron levels of subjects ($P < 0.05$); indeed, iron stores were improved as a result of beetroot juice intake. This result supports previous studies reporting that beetroot is an excellent source of iron (10). Whereas, in the control group, the levels of iron decreased after 6 weeks of soccer training ($P < 0.05$) which can be linked to their menstrual cycles (4) and soccer trainings (17). In compared to control group, MCV levels showed mild increases in 5 subjects in the experimental group. This may be related to the fact that 6 weeks of beetroot juice consumption was not sufficient for convincing results in MCV levels; this was consistent with the study conducted by Nora (15). Indeed, as the results of the study unfolded, there were no significant improvements in the MCV levels of subjects ($P > 0.05$) in the experimental group; there were also no significant improvements in MCV levels ($P > 0.05$) in the control group. In the experimental group significant improvement was observed in the ferritin levels of subjects ($P < 0.05$); this increase in ferritin may be related to the improvement of iron stores as a result of beetroot juice intake. Whereas, in the control group, levels of ferritin was decreased. In other words, there was a mild decrease in the ferritin levels of subjects ($P < 0.05$) that can be linked to their menstrual cy-

Table 2. Hb, Hct, RBC, Iron, MCV, Ferritin and TIBC Values Before and After Beetroot Juice Consumption in the Experimental and Control Groups

	Pre[a]	Post[a]	*t*	P Value
Hb (g/dL)				
BR	12.77 ± 0.44	14.21 ± 0.24	-10.85	0[b]
PL	12.87 ± 0.41	12.86 ± 0.47	0.31	0.75[c]
Hct (%)				
BR	39.06 ± 1.70	42.22 ± 0.69	-7.59	0[b]
PL	41.37 ± 1.18	40.55 ± 0.82	3.11	0.01[b]
RBC ($\times 10^6/\mu L$)				
BR	4.31 ± 0.16	5.11 ± 0.20	-11.80	0[b]
PL	4.46 ± 0.32	4.43 ± 0.34	1.08	0.30[c]
Iron ($\mu g/dL$)				
BR	79.50 ± 11.64	86.10 ± 11.69	-21.60	0[b]
PL	87.70 ± 15.46	86 ± 15.49	6.53	0[b]
MCV (fL)				
BR	91.92 ± 1.43	92.25 ± 1.71	-1.10	0.29[c]
PL	90.57 ± 3.28	90.56 ± 3.30	0.22	0.82[c]
Ferritin (ng/mL)				
BR	36.20 ± 2.28	37.09 ± 2.21	-5.28	0.001[b]
PL	36.58 ± 1.66	36.24 ± 1.67	12.75	0.000[b]
TIBC ($\mu g/dL$)				
BR	365.90 ± 28.42	351.10 ± 31.96	4.99	0.001[b]
PL	347.50 ± 29.61	340.60 ± 28.11	12.17	0.000[b]

Abbreviations: BR, beetroot; Hb, hemoglobin; Hct, hematocrit; MCV, mean corpuscular volume; PL, placebo; RBC, red blood cells; TIBC, total iron binding capacity.
[a]Values are expressed as mean ± SD.
[b]P < 0.05.
[c]P > 0.05.

Table 3. Comparison of Hematological Parameter Scores (Mean Difference) Between the Experimental and Control Groups

	Experimental[a]	Control[a]	*t*	P Value
Hb (g/dL)	1.44 ± 0.41	-0.01 ± 0.09	10.63	0.000
Hct (%)	3.16 ± 1.31	-0.81 ± 0.82	8.08	0.000
RBC ($\times 10^6/\mu L$)	0.80 ± 0.21	-0.02 ± 0.08	11.38	0.000
Iron ($\mu g/dL$)	6.60 ± 0.96	-1.70 ± 0.82	20.67	0.000
MCV (fL)	0.33 ± 0.96	-0.003 ± 0.04	1.11	0.28
Ferritin (ng/mL)	0.89 ± 0.53	-0.34 ± 0.084	7.21	0.000
TIBC ($\mu g/dL$)	-14.80 ± 9.36	-6.90 ± 1.79	-2.62	0.017

[a]Values are expressed as mean ± SD.

cles (4) and soccer training (17). In the experimental group, there was a significant decrease in TIBC levels ($P < 0.05$); this decrease in TIBC may be related to the improvement of iron stores as a result of beetroot juice consumption, whereas in the control group, the levels of TIBC decreased ($P < 0.05$).

4.1. Conclusions

Anemia in association with iron deficiency has serious implications (18). The earliest stage of iron deficiency is iron depletion. The most advanced stage of iron deficiency is iron deficiency anemia. It is characterized by decreased or absent iron stores, low serum iron and hemoglobin concentrations. Hemoglobin determination is one of the most convenient assessing methods in anemia (15). Beetroot is a source of substances and minerals. In this study, we recorded obvious increases in Hb, Hct, RBC, iron and ferritin levels after taking 200 mL beetroot juice for 6 weeks. This was consistent with the study conducted by Indhumathi and Kannikaparameswari stating that *B. vulgaris*

made significant increases in packed cell volume (PCV), hemoglobin concentrations, and red blood cell counts (RBCs) (19). Thus, it can be stated that the consumption of beetroot juice can be beneficial for maintaining good health and enhancing athletic performance, with some therapeutic properties for eliminating anemia in female soccer players. So, it is recommended that adolescent female soccer players put beetroot juice in their diet. A glass of red beetroot juice a day will not only provide the body with the required nutrients, but may also protect and prevent anemia.

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