

# Original Research Effects of natural iron supplement intake on EPO, Hp, and iron metabolism during rapid weight loss in university wrestlers

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#### Abstract

**Background**: The purpose of this study was to determine the changes in erythropoiesis and iron metabolism indicators in order to improve the maintenance of athletic performance during rapid weight loss. **Methods**: The subjects were 23 university wrestlers (13 male, 10 female). The first measurements in the male and female groups after natural iron supplement intake indicated weight decreases of 6.2% and 5.8%, respectively. After the 3-week washout period, the weights in the male and female groups decreased by 6.1% and 6.2%, respectively. **Results**: Males in the intake group experienced changes in iron metabolism after weight loss, as shown by increases in the iron indicators (Fe, p < 0.041; transferrin, p < 0.004; total iron binding Capacity (TIBC), p < 0.031), whereas the female group showed no significant changes. Both groups experienced decreases in erythropoietin (males, p < 0.021; females, p < 0.027) and VO<sub>2</sub>max (p < 0.001) after weight loss. Blood lactate in the intake group decreased after maximal exercise immediately after the test for the male group (p < 0.031) and during resting time and immediately after exercise in the female group (p < 0.001 and p < 0.050, respectively). **Conclisions**: These findings suggest that a 7% loss in weight does not positively affect athletic ability. Further research on methods for iron supplement intake, weight loss, and training is necessary to confirm our conclusions.

Keywords: rapid weight loss; athlete nutrition; exercise performance; iron supplements

## 1. Introduction

Wrestlers are organized into different weight classes during competition to ensure equitable matches between physically comparable athletes [1]. Wrestling is a weightdivision event requiring a combination of skill, dexterity [2,3], strength, and flexibility, in which many athletes undergo weight loss to compete. It has been reported that 80.6% of competitors are losing weight before each match [4,5]), since wrestlers have to have similar physical strength among their competitors to win [6,7]. Therefore, in order to win a match, wrestlers use weight loss as a method of improving athletic ability [8], and many wrestlers believe that weight loss affects this ability [9,10].

However, weight loss induces various physiological dysfunctions. According to studies by Imai *et al.* [11] and Sung *et al.* [10], a 5% rapid loss in weight negatively effects the aerobic ability, pro inflammatory, and blood lactate concentration of wrestlers. Weight reduction also induces losses of trace minerals and blood ions during prolonged exercise, including zinc and iron through sweat [12]. Iron is an essential mineral for delivering oxygen to body tissue as well as playing important roles in metabolism, respiration, and immune function. The body carefully maintains the iron level by ensuring a balance between lost, absorbed, and stored iron [13,14]. However, the excessive loss of blood ions (including iron) after intense training is

considered a major negative consequence of rapid weight reduction.

Physiological changes induced by intense training can mimic iron deficiency and decrease the blood concentrations of hemoglobin (Hb) and ferritin [15,16]. Iron metabolism disorder has causes besides erythrocytosis, one of which is a decrease in haptoglobin (Hp). Hp is a plasma protein synthesized by the liver and released into the circulation, and plays an important role in iron metabolism [17]. Hp is the marker of erythrocytosis and combines with Hb to prevent Hb loss and prevent the loss of iron in Hb [18,19]. Hp combines with damaged Hb to prevent it from losing iron.

Based on the above-described situation, we attempted to determine the effect that natural iron supplementation has on the endurance capacity and erythropoiesis in wrestlers participating in rapid-weight-loss programs. These preliminary results can serve to generate further hypotheses and facilitate plans for using iron supplementation in nutritional protocols combined with exercise programs for wrestlers. We hypothesized that difference between the level of rapid weight Loss with natural iron supplements affects VO<sub>2</sub>max, blood lactate concentration, and iron metabolism.



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## 2. Materials and methods

## 2.1 Participants

The test subjects were 23 wrestlers (13 males and 10 females) over six years of wrestler in the Korean Olympic Committee from Yongin University and Sohea College. All subjects received a sufficient explanation of the research content, purpose, and associated procedures prior to participation and voluntarily consented to participate in the experiment. All participants trained, ate, and slept at the same time, and ate the same menu. In addition, drinking, smoking, and taking supplements during the experimental period were restricted.

#### 2.2 Experimental design

The study consisted of a single-blind test with two trials, including a 3-week washout period, labeled trial 1 and trial 2 (Fig. 1). The study consisted of a single-blind test with two trials, including a 3-week washout period, labeled trial 1 and trial 2. Each experiment consisted of the same participants (13 males and 10 females), in the first experiment, they lost weight by taking iron supplements, and in the second experiment, they washed out for three weeks and then supplemented placebo to lose weight. Trial 1 consisted of a first-day pretest (body composition, blood sampling, and a maximal incremental running test) followed by a 7% weight loss over 7 days while consuming a natural iron supplement. The participants were instructed to drink the iron supplement (Spatone water, Nelsons, UK) (Table 1) containing 5 mg of iron and orange juice at 6:00 and 20:00 (two pouches per day), for the week-long weight-loss period. Trial 2 consisted of a pretest, followed by an approximately 7% weight loss over 7 days only drinking orange juice. A posttest that followed the same procedure as the pretest was conducted on day 7 at 6:00 for both trials. During the weight-loss period, subjects completed the training protocol two or three times each day, totaling around 5 hours of exercise each day, and 30 hours over the week. Also during the washout period, participant controlled exercise and diet.

Table 1. Nutritional composition of iron supplement.

Serving size	Amount per serving	% Daily value		
Kcal	0 kal			
Carbohydrate	0 g	0%		
Protein	0 g	0%		
Fat	0 mg	0%		
Sodium	0 mg	0%		
Iron	5 mg	42%		

## 2.3 Weight reduction method

For this study, it was important to develop the training program to reduce the body weight since we focused

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only on exercise intensity without dietary regulation. The method of losing weight was through the same diet and exercise program. Diet supplements were completely banned, but additional food intake was not controlled. As such, the core strategies for the training program were for the wrestlers to lose body weight and maintain their athletic performance. Therefore, the types of included exercises were intended to achieve explosive strength, muscle strength, muscle endurance, and cardiovascular endurance. For the morning session, the main training consisted of running and dashing to increase cardiovascular endurance and muscle endurance. The dash training for explosive strength was performed on a plain, a hill, and a stair on Monday, Wednesday, and Thursday. The interval training and fartlek training were performed for cardiovascular endurance and muscle endurance on Tuesday and Friday. To reinforce insufficient training, subtraining was completed after each main training session. For the afternoon session, mat training was conducted targeting physical fitness, skill training, sparring, and ground sparring to improve game strategy. For the evening session, the subjects performed weight training to maintain their strength on Tuesday and Thursday, but took a rest to recover on Monday, Wednesday, and Friday. The weight training program consisted of a power clean, dead lift, squat, and sit-ups to exercise the power zone, including the lower back, abdominal region, and thighs

### 2.4 Maximal incremental running test

In this study, subjects participated in a maximal incremental running test on a treadmill (quasar 4.0, h/p/cosmos sports & medical gmbh, Traunstein, Germany) using the Bruce protocol [20]. The subjects arrived 1 hour before the experiment, when we measured their respiration rate, ventilation volume, and respiratory exchange ratio. After warming up at a self-selected speed, the maximal incremental test commenced, beginning with 2 min at the current speed. During the test, subjects were monitored using ECG, and stopped when they appeared to have continuous ventricular tachycardia, decreases of >1 mm of the ST segment without a leading diagnostic Q-wave, or volitional exhaustion despite encouragement from the researcher.

#### 2.5 Blood sampling and analysis

Blood samples were collected from the brachial vein four times (during each trial's pre- and post-test: before body composition measurement). Plasma aliquots were stored in a freezer after centrifugation for subsequent analysis. All eight types of blood cells, including erythropoietin (EPO) and Hp, and the iron metabolism index were analyzed by Green Cross Laboratories in Korea.

#### 2.6 Blood lactate analysis

Blood samples were collected from the brachial vein four times (during each trial's pre- and post-test: before

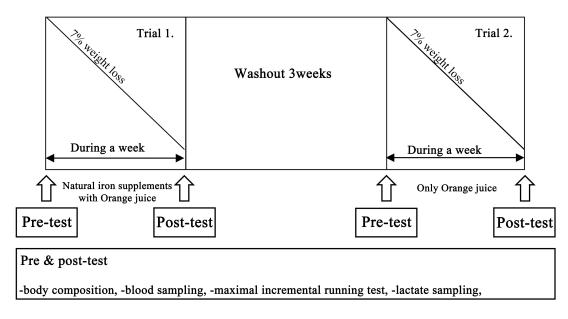


Fig. 1. The experimental design.

body composition measurement). Plasma aliquots were stored in a freezer after centrifugation for subsequent analysis. All eight types of blood cells, including erythropoietin (EPO) and Hp, and the iron metabolism index were analyzed by Green Cross Laboratories in Korea.

#### 2.7 Statistical analysis

Statistical analyses were conducted using SPSS (version 25.0 for Windows, IBM Corp., Chicago, IL, USA). All data were presented using mean  $\pm$  SD values. All groups checked the normality of data, two-way repeated-measures ANOVA with post-hoc tests by paired *t*-tests was then used to identify the differences in blood parameters between the pre- and post-test after consuming a natural iron supplement. Statistical significance was set at p < 0.05.

## 3. Results

## 3.1 Participant body weight changes

Table 2 lists the basic characteristics and the changes following natural iron supplementation of the participants, including their age, height, and body weight and composition. The body weights for the male group were 73.2  $\pm$  7.1 kg and 73.7  $\pm$  6.7 kg in the iron-supplementation and placebo groups, respectively. After the rapid-weight-loss program, the body weights decreased by 6.2% and 6.1% in the iron-supplementation and placebo groups, respectively. For the females, the body weights were 65.2  $\pm$  7.7 kg and 67.8  $\pm$  6.9 kg in the iron-supplementation and placebo groups, respectively, which decreased by 5.8% and 6.2% after the rapid-weight-loss program. There were no significant body weight changes between the first and second trials over the 3-week washout period, indicating that our weight-loss program was effective in both trials.

### 3.2 Changes in hematological factors

Table 3 lists the changes in hematological factors. For males there was no significant increase in red blood cells (RBCs) in the iron-supplementation group, while they a decrease in the placebo group after the rapid-weightloss program. Hb and hematocrit (Hct) increased significantly in the iron-supplementation group (p < 0.022 and p < 0.002, respectively), but decreased in the placebo group. For females, Hct increased significantly in the ironsupplementation group (p < 0.004), as did RBCs (from 4.48  $\pm$  0.22 to 4.58  $\pm$  0.30). Changes in the other blood cell types (Mean corpuscular volume (WCV), platelets, While cell volume (MCH), and Mean corpuscular hemoglobin concentration (MCHC)) were not significant for both males and females after the rapid-weight-loss program. The blood iron concentration increased in both males and females. Blood iron increased in the iron-supplementation group (by 20.53% and 19.41% in males and females, respectively) and placebo group (4.73% and 13.71%) with no significantly. Only being significant in males (p < 0.041). In the male group, both transferrin (p < 0.001) and TIBC (p< 0.001) increased, with significant changes in the ironsupplementation group. The iron metabolism index (transferrin, ferritin, and TIBC) was no significantly in females after the rapid-weight-loss program. Lastly, Hp was not significantly change in either males or females after the rapidweight-loss program.

#### 3.3 Changes in EPO levels

EPO decreased after rapid weight loss in males from  $14.94 \pm 6.86$  to  $9.51 \pm 4.03$  and from  $12.48 \pm 3.14$  to  $9.21 \pm 3.04$  (p < 0.021) in the iron-supplementation and placebo groups, respectively; the corresponding decreases

Table 2.	Basic	characteris	tics of	partici	pants.
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	Male				Female			
Factor	Iron		Placebo		Iron		Placebo	
	Pre	Post	Pre	Post	Pre	Post	Pre	Post
Age (years)		21.6	$\pm 0.8$		20 ± 1			
Height (cm)	$169.8\pm4.7$				$162.4 \pm 6$			
Body weight (kg)	$73.2\pm7.1$	$68.3\pm 6.2$	$73.7\pm6.7$	$69.3\pm6.1$	$65.2\pm7.7$	$61.4\pm7.3$	$67.8\pm6.9$	$63.7\pm9.5$
SMM (kg)	$35.4\pm3.8$	$33.3\pm3.1$	$34.9\pm3.8$	$33.6\pm2.7$	$25.8\pm2.3$	$24.5\pm2.1$	$27.1\pm1.7$	$25.8\pm2.9$
Fat mass (kg)	$12.3\pm2.9$	$9.8 \pm 1.9$	$12.7\pm3.1$	$10.1\pm2.1$	$18.1\pm3.9$	$16.7\pm3.9$	$19.6\pm4.1$	$17.1\pm4.6$
Body fat (%)	$16.2\pm2.7$	$14.6\pm2.2$	$17.5\pm2.8$	$14.2\pm2.3$	$27.2\pm2.9$	$26.2\pm2.9$	$28.6\pm3.2$	$26.7\pm3.5$
$BMI (kg/m^2)$	$25.7\pm2.1$	$24.1\pm1.2$	$25.5\pm2.1$	$24.4\pm1.2$	$24.7\pm2.3$	$23.6\pm1.9$	$25.9\pm2.2$	$24.3\pm2.7$

Values expressed as mean  $\pm$  SD.

SSM, Skeletal muscle mass; BMI, Body mass index.

in females were from  $14.45 \pm 6.07$  to  $12.53 \pm 4.04$  and from  $11.83 \pm 2.38$  to  $9.45 \pm 1.83$  (p < 0.027), respectively.

#### 3.4 Changes in endurance capacity based on VO<sub>2</sub>max

VO<sub>2</sub>max decreased in males from 49.81  $\pm$  2.78 to 45.51  $\pm$  2.07 (-8.7%) and from 48.02  $\pm$  2.45 to 43.99  $\pm$  2.63 (-9.4%) after rapid weight loss in the iron-supplementation and placebo groups, respectively. The iron-supplementation group showed smaller decreases than the placebo group with statistically significant (p < 0.001). VO<sub>2</sub>max decreased in females from 48.91  $\pm$  2.91 to 44.72  $\pm$  1.37 (-8.6%) and from 48.36  $\pm$  2.84 to 44.12  $\pm$  2.01 (-8.8%) in the iron-supplementation and placebo groups, respectively (p < 0.001) (Fig. 2).

#### 3.5 Changes in blood lactate levels

Among males immediately after testing, blood lactate levels decreased in both groups, but with no significance. After 10 min of recovery, blood lactate levels in the iron-supplementation group had significantly decreased (p < 0.031). For males during the resting period, the placebo group had significantly higher blood lactate levels after rapid weight loss (p < 0.001). Immediately after testing, males in the iron-supplementation group showed a significant decrease in blood lactate levels after rapid weight loss (p < 0.050) (Table 4).

#### 4. Discussion

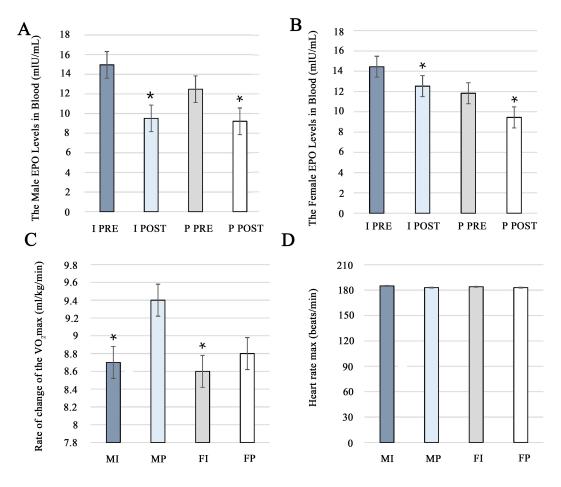
We hypothesized that difference between the level of rapid weight Loss with natural iron supplements affects  $VO_2max$ , blood lactate concentration, and iron metabolism. And, results have shown that there is a difference between blood lactate concentration and iron metabolism. However, differences in  $VO_2max$  could not be identified depending on the rapid weight loss level.

It is already well known that short-term weight loss of around 5% does not affect physical fitness. Sung *et al.* [10] found that university wrestlers taking natural iron supplements during weight loss did not significantly affect their VO<sub>2</sub>max. This supports our result of no changes in physical strength with a 5% weight loss. However, weight loss of 7% in the present study had a negative effect on the aerobic exercise capacity of wrestlers. Our results on the changes in blood lactate concentrations during the recovery period were similar to those in the study of Franchini *et al.* [9], which applied a 5% weight loss. But the changes in aerobic capacity be attributed to cumulative training fatigue and not weight loss. Because participants have a 30-hour training week and maximal test pre and post with an aggressive weight loss protocol. This could be due to glycogen depletion and central fatigue [21–23].

The recovery speed of maximum blood lactate concentration during postweight and placebo administration was faster, resulting in a significant interaction effect during the 10-min recovery. The iron-supplementation group tended to show significant decreases in blood lactate when resting before maximal exercise and during the 10 min of recovery, especially in females. This result suggests that iron intake increases the efficiency of muscle contractions. Our study also suggests that low lactate levels were due to the low maximum oxygen uptake resulting from the low contribution of maximal exercise performance.

Also did not use nonhems and soluble hems, which are commonly used, but instead induced rapid absorption of natural liquid iron, to help recover from peripheral fatigue and improve oxygen transport as indicated by Hinton *et al.* [24] and Sung *et al.* [10]. The 7% weight loss did not confirm the positive effects of natural iron supplementation on maximum oxygen uptake, and further research on the duration and dosage of iron ingestion is therefore needed to confirm this possibility. But to link iron metabolism to reductions in weight loss then you need to consider the research on change in iron status with low energy availability.

We also examined the changes in blood iron content between before and after weight loss, and found the same concentration of iron after weight loss as in the study of Sung *et al.* [10]. Our study indicated that on average, blood iron concentration after iron supplementation during weight loss increased compared with that in the placebo. Similar



**Fig. 2. Effect of natural iron supplement no the exercise capacity.** (A) The EPO Levels in Blood (Male). (B) The EPO Levels in Blood (Female). (C) Rate of change of the VO<sub>2</sub>max. (D) HRmax. I PRE, Iron pre-test; I POST, Iron post-test; P PRE, Placebo pre-test; P POST, Placebo post-test; MI, Male iron; MP, Male placebo; FI, Female iron; FM, Female placebo.

research has found that blood iron concentration increased after weight loss compared with before, as the ions released from the intracellular fluid increased the concentration of extracellular fluid [25,26]. Previous studies have shown these results and as well as significant results associated with 7% weight loss.

Finally, changes in EPO and Hp were expected to increase RBC production from natural iron ingestion, conserving the aerobic capacity reduced by weight loss. However, EPO tended to significantly decrease immediately after weight loss for all of the present study groups. Hp also showed no significant differences. Changes in Hp and Hb are largely indicators of haemolysis in athletes that may be seen immediately after a training session of prolonged duration or intensity [27]. According to Smith et al. [28], the increase in erythrocytosis due to weight loss demands excessive training to reduce Hb in the body, and to promote production of EPO. In our study the RBC and Hb levels after iron supplementation also increased significantly compared with the placebo group, whereas EPO showed no significant differences between the intake and placebo groups. If EPO levels were affected by RBC or Hb levels, EPO levels in the iron-supplementation group should have increased, which we believe was affected by the 7% weight loss, which is an abnormal reduction over a 1-week period. In the initial adaptation to training where we see sport anemia resulting in the dilution of Hb which is the iron deficiency mimicking.

Clapp et al. [29] reported that EPO showed an aboveaverage increase after exercise. A prior study suggested that decreases in EPO and Hp are also linked to the production of RBCs, but due to the abnormality of the 7% weight loss, the correlations between natural iron supplementation and EPO and Hp levels is inconclusive. Further studies using different weight-loss periods are therefore needed. Sandström et al. [30] and DellaValle et al. [14] suggested that Hp changes should decrease after combining with Hb, but we found no significant differences. The abnormal weight loss and exercise could also be factors, highlighting the need for further studies of EPO, Hp, iron metabolism, and weight loss. Besides, The reason why the difference in Hb after 7% weight loss is not large is that weight loss was performed for a short period of time using RWL. Viveiros et al. [31] reported that there was no change in sodium, potassium,

		Ma	le		Female			
Factor	Iron		Placebo		Iron		Placebo	
	Pre	Post	Pre	Post	Pre	Post	Pre	Post
WBC (10 <sup>6</sup> /lL)	$6.64\pm0.12$	$7.28 \pm 1.05$	$7.18 \pm 1.67$	$6.56 \pm 1.34$	$7.49 \pm 0.86$	$8.36 \pm 1.41$	$7.89 \pm 2.02$	$6.74\pm0.43$
RBC (10 <sup>6</sup> /lL)	$5.11\pm0.12$	$5.26\pm0.13$	$5.32\pm0.12$	$5.06\pm0.23$	$4.48\pm0.22$	$4.58\pm0.30$	$4.91\pm0.35$	$4.82\pm0.36$
Hb (g/dL)	$15.61\pm0.39$	$\textbf{16.03} \pm \textbf{0.39}^{*}$	$16.28\pm0.66$	$15.30\pm0.72$	$14.36\pm0.43$	$13.88\pm0.62$	$14.78\pm1.43$	$13.96 \pm 1.12$
Hct (%)	$47.21 \pm 1.22$	$\textbf{48.13} \pm \textbf{1.25}^{**}$	$49.18 \pm 1.4$	$47.3\pm1.5$	$44.74 \pm 1.37$	$\textbf{46.38} \pm \textbf{2.52}^{**}$	$45.37\pm3.75$	$44.99 \pm 2.65$
Platelet (10 <sup>3</sup> /lL)	$300.69\pm34.23$	$316.76\pm42.88$	$309.53\pm46.39$	$294.92\pm59.72$	$339.1\pm32.99$	$306.8\pm52.71$	$295.6\pm50.09$	$2982.5\pm29.46$
MCV (fL)	$92.29 \pm 2.45$	$91.63 \pm 2.44$	$92.46 \pm 2.06$	$91.33 \pm 2.19$	$92.71 \pm 2.28$	$91.63 \pm 2.44$	$93.21 \pm 2.37$	$92.19 \pm 1.43$
MCH (pg)	$30.43\pm0.93$	$30.51\pm0.89$	$30.63 \pm 1.10$	$29.54 \pm 1.23$	$30.14 \pm 1.16$	$30.41\pm0.93$	$30.56 \pm 1.22$	$29.01 \pm 1.17$
MCHC (%)	$33.08\pm0.29$	$33.28\pm0.66$	$33.11\pm0.59$	$33.35\pm0.9$	$32.42\pm0.65$	$32.87\pm0.7$	$32.77\pm0.75$	$31.46\pm0.9$
Fe (µg/dL)	$81.92 \pm 14.06$	$\textbf{103.08} \pm \textbf{25.31}^*$	$117.30\pm33.11$	$121.84\pm49.81$	$87.2\pm24.15$	$108.2\pm33.93$	$98.21 \pm 39.57$	$113.8\pm49.81$
Transferrin (mg/dL)	$255.51\pm14.08$	$\textbf{322.15} \pm \textbf{18.59}^*$	$285.4\pm23.11$	$332.84\pm27.27$	$285.61\pm42.61$	$292.94\pm31.43$	$276.79\pm27.58$	$308.09\pm45.65$
Ferritin (ng/dL)	$146.77\pm28.58$	$143.54\pm45.13$	$120.61\pm50.03$	$148.23\pm 66.12$	$116.01\pm53.48$	$126.1\pm60.13$	$93.70\pm61.65$	$132.60\pm51.91$
TIBC (µg/dL)	$321.92\pm18.76$	$\textbf{322.15} \pm \textbf{18.58}^*$	$345.92\pm26.13$	$284.33\pm23.25$	$347.7\pm49.86$	$328.01\pm43.19$	$338.2\pm33.94$	$326.04\pm52.75$
Ip (mg/dL)	$29.38\pm12.53$	$30.54 \pm 9.96$	$50.53 \pm 18.52$	$33.39 \pm 12.16$	$40.02\pm21.51$	$39.31 \pm 15.65$	$63.5\pm21.24$	$44.11\pm14.28$

Table 3. Complete blood cell types, iron and haptoglobin levels in blood.

Values expressed as mean  $\pm$  SD. \*: p < 0.05, \*\*: p < 0.01 vs Pre by *t*-test. Bold data indicate the significance.

WBC, White blood cell; RBC, Red blood cell; Hb, Hemoglobin; Hct, Hematocrit; MCV, Mean corpuscular volume; MCH, While cell volume; MCHC, Mean corpuscular hemoglobin concentration; Fe, Iron; TIBC, Total iron binding Capacity; Hp, Haptoglobin.

Table 4. Lactate levels in blood.								
Group			Resting	IAT	10 min	30 min		
	Iron	Pre	$0.99\pm0.14$	$17.81 \pm 1.93$	$5.56\pm0.72$	$2.15\pm0.64$		
Male		Post	$1.01\pm0.13$	$18.01 \pm 1.91$	$\textbf{6.11} \pm \textbf{0.95}^*$	$2.61\pm0.68$		
Male	Placebo	Pre	$1.09\pm0.21$	$18.02 \pm 1.68$	$5.49\pm0.87$	$2.29\pm0.59$		
		Post	$1.08\pm0.11$	$17.84\pm2.16$	$6.54 \pm 1.01$	$2.54\pm0.66$		
	Iron	Pre	$0.96\pm0.15$	$16.12\pm2.09$	$6.59\pm0.82$	$3.01\pm0.69$		
Female		Post	$\textbf{0.94} \pm \textbf{0.14}^{***}$	$16.72\pm1.80$	$\textbf{7.03} \pm \textbf{0.81}^*$	$3.28\pm0.55$		
remale	Placebo	Pre	$0.97\pm0.15$	$16.30\pm2.35$	$6.55\pm.084$	$3.42\pm0.57$		
		Post	$1.07\pm0.35$	$16.19\pm2.34$	$7.55\pm0.85$	$3.11\pm0.059$		

Values expressed as mean  $\pm$  SD. \*: p < 0.05, \*\*\*: p < 0.001 vs Pre by *t*-test. Bold data indicate the significance.

IAT, Immediately after test; 10 min, After recovery 10 min; 30 min, After recovery 30 min.

chloride, hematocrit, and hemoglobin levels even if rapid weight loss was performed on male and female wrestlers. Since RWL was used in this study, I think a conclusion similar to that of previous studies was drawn.

The limitations of this study include that the number of participants was small and limited to wrestlers. Inaddition, the experiment lasted only over a week, and there was a lack of analysis of the related factors of iron metabolism, including hapsidin, and controlled the diet of wrestlers, but no additional food intake was controlled. The method of weight reduction also use identical exercise program, but there was no control over the method of weight loss (Sauna, individual training) other than exercise. Finally, measurement on Erythroferrone and regulation of hepcidin and iron status would be required.

## 5. Conclusions

The purpose of study was to investigate the changes in the EPO, Hp and iron metabolism indicators for an enhancement of the health maintenance and improvement of athletic performance during a short-term weight loss University Wrestlers. In summing up the results of this study, both male and female athletes have seen their EPO levels decrease, Hp has not changed with a significant after about 7% weight loss. For EPO and HP, the significant static increase was expected from the intake group after weight loss in the natural iron supplement intakes, but we found that around 7% of weight loss was not positively affected. In addition, aerobic exercise ability was also negatively affected, but we found out that natural iron supplement intakes positively affected the blood lactic acid after a maximal incremental running test. Also, the results show that iron metabolism for the male group the potential for iron effects was identified due to significant static increases such as Hb, Hct, and iron metabolism indicators for iron deficiency and red blood cell damage that can occur during short-term weight loss. But in the female group, the significant effects of taking natural iron tablets could not be identified. It is considered that because the female athletes have a higher iron loss factor than male athletes, which has a more negative effect during weight loss. Lastly, highlighting the need for further studies of the method of iron supplement intakes, weight loss and training, additional studies related to dietary intake will be needed and further research regarding the amount and timing of iron supplements.

## Abbreviations

Nat, the national team; Sen, the senior team; Uni, the university team; HDL-C, high-density lipoprotein-cholesterol; LDL-C, low-density lipoprotein-cholesterol.

## **Author contributions**

JYS, TWO and SGL provided funding. TWO conceived and performed experiments. JYS drafted the manuscript and provided expertise and feedback. SGL conceived and performed experiments.

## Ethics approval and consent to participate

This study was conducted according to the guidelines laid down in the Declaration of Helsinki andall procedures involving human subjects were approved by the research ethics committee at the YongIn University (approval no. 2-1040966-AB-N-01-20-1803-H MR-0992). Written consent was obtained from all subjects.

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## **Conflict of interest**

The authors declare no conflict of interest.

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