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# The Impact of 4% Rapid Weight Loss on Leptin, Adiponectin, and Insulin Resistance Among Elite Adult Freestyle Wrestlers

Mohammad Talaei,<sup>1</sup> Farzad Nazem,<sup>1</sup> and Mohammad Taghi Goodarzi<sup>2</sup>

**ABSTRACT.** Rapid weight reduction techniques that emphasize severe restrictions of food intake and water consumption during a short period of time are commonly employed by wrestlers before weigh-in. Along with the negative effects of rapid weight loss on a wrestler's physiological functions, we investigated leptin and adiponectin levels and insulin resistance in young wrestlers during their rapid weight loss program. Fifteen freestyle wrestlers were randomly selected as the subjects. They had a mean of age  $23 \pm 1$  y and anthropometric characteristics of: weight  $67.6 \pm 0.8$ , BMI  $22.5 \pm 0.21$  kg/m<sup>2</sup>, body fat percentage  $6.12 \pm 0.18$ , waist-to-hip circumference ratio  $0.82 \pm 0.08$ . Caloric intake (mean 7 days measured by food analyzer software) and anthropometric characteristics were measured by standard methods. The concentrations of the leptin and adiponectin hormones and insulin resistance index were measured with a sandwich ELISA kit method and (HOMA) from fasting glucose and insulin levels, respectively. Rapid weight loss program with a 4% weight loss had a significant impact on anthropometric factors, with decreasing leptin level, insulin resistance, and increased beta cell function, while the levels of adiponectin did not significantly change after weight loss. Rapid weight loss has harmful physiological effects on wrestler's bodies, but can be associated with improvements in the regulation of fatty acid, glucose metabolism, and insulin resistance.

**Keywords:** freestyle wrestling, insulin resistance (HOMA-IR), leptin-to-adiponectin ratio, rapid weight loss

## INTRODUCTION

Wrestling, along with other combative sports such as taekwondo, boxing, judo, as well as weightlifting, are weight-regulated sports that require athletes to achieve the weight of their desired category prior to competition. Both styles of international wrestling, Greco-Roman and freestyle, are performed with two rounds of 3 minutes with a 30-second-interval rest between rounds. The nature of wrestling demands that techniques be performed frequently with high, or close to maximum, intensity during a short period of time and imposes high metabolic stress on the athletes. The efficiency of the glycolytic and oxidative energy systems, muscular en-

durance and power, and lactate tolerance are very important to optimum athletic performance (Kraemer & Janson, 2004). A muscular body composition (mesomorph) in weight-regulated sports can provide an anthropometric and biologic advantage in performing specific athletic skills. Therefore, wrestlers attempt to manipulate their weight and body size in order to maximize their strength and power-to-weight ratio to compete with their opponents (Kordi & Rostami, 2011). Adipose tissue in wrestlers has been shown to be 10–12% during the off-season, with reductions to 4–6% during in-season (Carger & Crawford, 1992).

The phenomenon of rapid weight loss is also common among elite wrestlers in different age groups. Many wrestlers hope to increase their chance to be successful by making weight in one or two weight classes lower than their actual weight. Although rapid weight loss, according to coaches' opinions, is an ergogenic aid, sometimes the athlete's health is neglected (Lambert & Jones, 2010; Roemmich & Sinning, 1997; Viscardi, 2005). The findings of Balban (1989) and Alderman et al. (2004) showed that wrestlers lose their weight by various methods including severe caloric and wa-

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ter restriction, intense dehydration using saunas, performing exercise in warm environments, and use of diuretic and expellant drugs. Consequences of the applications of these methods lead to depletion of energy stores, decreased body water and electrolyte levels, muscle mass loss, and eventually, disorders in optimum physical function and athletic performance. The detrimental effects of rapid weight loss such as depletion of energy stores, decreased muscular strength and endurance, decreased aerobic capacity and anaerobic power, mental confusion, weakness, irritability, malnutrition, eating and nutrition disorders, and even death are well documented (Perriello, 2001; Tarnopolsky, 1996; Watson & Judelson, 2005).

In recent decades, the traditional view on the role of adipose tissue as bio-insulating and triglyceride storage has changed. Current studies demonstrate that specific proteins known as adipocytokines are synthesized from adipose tissue. These polypeptide hormones such as leptin and adiponectin are involved in the metabolic profile, appetite balance, insulin sensitivity, immune function and the onset of cardiovascular and diabetes diseases (Trayhurn, 2007; Rexford, 2006), and on the other hand excessive adipose tissue is a health risk factor, as well as having the potential to decrease performance in weight-regulated sports (Wagner, 1996). Leptin is a peptide hormone with a 167-amino acid sequence and a 16 KD molecular weight and is exclusively secreted by adipose tissue. It regulates energy balance and metabolism and is involved in controlling body weight. Leptin also has effects on the central nervous system, particularly the hypothalamus, by decreasing food intake and increasing energy expenditure. Increased leptin concentration in the hypothalamus arcuate nucleus decreases the production of ARP (agouti-related protein) and Y neuropeptide hormone, and simultaneously increases the function of anti-appetite hormones (proopiomelanocortin and cocaine- and amphetamine-related transcript CART). Eventually these factors cause a decreased appetite. Leptin also increases the beta-oxidation rate of fatty acids that are stored in the muscles, but increased leptin resistance can result because of the inability of leptin to pass the blood-brain barrier, or a decrease in the number of its receptors. The amount of body fat stored regulates the basic level of leptin and an increase in the number of fatty cells is associated with an increased basal leptin concentration. Leptin levels are positively correlated with body mass index. Recent evidence reveals that a high baseline level of leptin may be associated with increased body weight and fat storage. The role of hormonal factors in the regulation of plasma leptin levels, such as cortisol, which stimulates leptin gene expression and is associated with increased leptin secretion from adipose tissue, has been demonstrated. Leptin levels are also dependent on dietary fat intake. Generally, the two factors that effectively influence basal leptin concentration are caloric restriction and regular physical activity, and are therefore associated with weight loss (Friedman, 2002; Myers & Cowley, 2008). Adiponectin

with a 248-amino acid sequence is known as an anti-diabetic and anti-inflammatory factor. The secretion of adiponectin is controlled exclusively by adipose tissue. It is thought that it has a role in the regulation of energy metabolism in the liver and in muscle tissues by stimulating free fatty acid consumption and hepatic glucose uptake. Unlike other adipocytokines, the baseline levels and gene expression of adiponectin can be decreased in obese bodies; in diabetic and cardiovascular patients it can be significantly increased by weight loss. The baseline concentration of adiponectin has an inverse relationship with body mass index, body fat percentage, waist circumference, fasting insulin, triglycerides, and LDL-C plasma levels (Assaad, 2008; Matsuzawa et al., 2004). Insulin resistance index, known as (HOMA-IR), is another factor that has an important role in decreasing the sensitivity of target tissues in response to insulin hormone action. Despite an increase in blood glucose (hyperglycemia), pancreatic beta cells must secrete more insulin to keep blood sugar in the normal range. Increased body weight stimulates more insulin secretion and an increase in insulin resistance, but decreased body weight associated with restriction of food intake and aerobic activity will enhance insulin sensitivity. Increased insulin secretion will decrease adiponectin concentrations and stimulate leptin gene expression, and all of these factors will lead to an increase in insulin resistance and a decrease in beta cell function (Ikeoka et al., 2010; Ferrannini et al., 2004). The findings from similar studies have shown inconsistent results. While Rolland et al. (2011) showed that a 5% weight loss through a 3-month caloric restriction had a significant reduction on leptin and an enhanced adiponectin levels in line with the present results, Mäestu et al. (2008) reported that a 6% loss of body weight by caloric restriction during the 10 weeks of the weight loss program had a significant decrease in the leptin level without considerable changes in the adiponectin concentration. On the other hand, weight loss before and during a formal tournament has an important role on the success of wrestlers. Restriction of food intake as well as water consumption influenced anthropometric variables and adipocytokines secreted from adipose tissue will transform the organic health and the process of glucose and lipid metabolism (Ransone and Hughes, 2004). Due to the lack of scientific research of this topic with athletes, the multiple roles of leptin and adiponectin within the rapid weight reduction of wrestlers was investigated.

## METHODS

This study was carried out using a causal-comparative design. Fifteen wrestlers with a weight ranging from 60–66 kg, age:  $23 \pm 1$ , and 7 years athletic experience were randomly selected from among wrestling clubs. The subject's medical histories were investigated for any cardiovascular or metabolic disease. All subjects were asked not to drink alcohol,

TABLE 1 Baseline Anthropometric Characteristics of the Wrestlers at Pretest (One Week Before Beginning Weight Loss Program)

<i>Variables</i>	<i>Pretest (n = 15)</i>	<i>Min–Max</i>
Age (Year)	23±1	20–26
Body Weight (kg)	68±0.8	64–71.5
Height (cm)	173	178–170
Body Mass Index (kg/m <sup>2</sup> )	22.5±0.2	21–23.8
Body Fat Percentage (%)	6.12±2.2	5.10–7.13
Waist Hips Ratio (cm)	0.82±0.08	0.77–0.88

smoke, or use any medication. They also went through a medical examination and gave a written informed consent. All details of the study were explained to the subjects and the study was approved by the ethics commission. Data for the anthropometric characteristics including height, weight, subcutaneous fat percentage, BMI, and waist-to-hip ratio were measured by the standard methods and are presented in Table 1. Anthropometric data and blood samples were collected at three stages, a week before the weight loss program, and 12 and 24 h after weigh-in. Blood samples were collected in heparinized tubes from the antecubital vein of the right hand in a fasting state and immediately stored in ice. Plasma was separated from cells by centrifugation at 3000 rpm for 10 minutes and the plasma samples were frozen at  $-80^{\circ}\text{C}$  until analysis.

### Weight Loss Program

Following the measurement of anthropometric characteristics and biochemical variables during the pretest stage, the wrestlers engaged in a weight loss program for 1 week through caloric restriction ( $1700\pm 100$  kcal/day) and water ( $1100\pm 95$  ml/day). Through this weight loss program the wrestlers were able to decrease 4% of their body weight. Wrestlers were not allowed to use saunas, rubber clothes, intense exercise in a warm environment, diuretic, or laxative drugs.

### Anthropometric Measurement

The body height of subjects was measured with a metal scale with 0.5 cm sensitivity, and body weight measurement was taken with lightweight clothing and without shoes with a digital scale with 0.1 kg sensitivity. Waist-to-hip ratio (WHR) was calculated as waist circumference in centimeters divided by hip circumference in centimeters. Body mass index (BMI) was calculated as weight (kg) divided by height (m) squared. Skinfolds were measured with a Harpenden mechanical caliper and entered into a six-site Tipton linear regression equation. All of the measurements were done after 8 h of overnight fasting, after eating breakfast, and emptying gastric waste.

### Daily Caloric Intake

The subjects kept an eating diary before and during the participation in the weight loss program listing all food consumed. Based on this, the daily energy intake was calculated as the average of 7 days by food analyzer software that was approved by the qualifiers of the Ministry of Health and under the control of expert dietitians.

### Biochemical Variables

Analyses of serum leptin and adiponectin was determined by the ELISA method with the sensitivity of 0.2 and 0.6 (ng/dl), respectively. A leptin-to-adiponectin ratio was taken as leptin divided by adiponectin. Fasting insulin was measured with a commercial kit (Monobind©, Lake Forest, California, USA) with a sensitivity of 0.75 (ng/dl). Fasting glucose levels were obtained with a glucose oxidase kit calorimetric method with a sensitivity of 0.2 mg/dl. The homeostasis model assessment (HOMA) was used to calculate insulin resistance and beta cell function in vivo according to the formulas presented by Matthews et al. (1985): fasting plasma glucose (mg/dl)  $\times$  fasting plasma insulin ( $\mu\text{u/liter}$ )/22.5 for insulin resistance and  $\text{insulin} \times \text{glucose}/20/3.5$  for beta cell function. To calculate insulin resistance and beta cell function, HOMA calculator software was used (Finucane, 2009). Statistical analyses were performed by SPSS version 18. Results of descriptive statistics in this study are presented as mean, standard error, minimum, and maximum values. Kolmogorov-Smirnov (KS) and Leven tests were used to determine normally distributed data and variance comparison, respectively. One-way ANOVA (repeated measures) was used in order to compare dependent parameters in three stages (pretest, 12h, and 24h recovery times) of the weight loss program. Statistical significance was accepted at ( $p < 0.05$ ).

## RESULTS

The findings of this study shown that the 4% of body weight reduction in wrestlers had a significant reduction on anthropometric variables (weight, BMI, body fat percentage, WHR) and are shown in Table 2. There was also a significant reduction in both the absolute and relative values of leptin and are listed in Table 3. Interestingly, the changes in adiponectin serum were not significant, while the value of adiponectin-to-body fat percentage changes showed a significant decrease after the rapid weight loss program and are shown in Table 4.

### Leptin-to-Adiponectin Ratio

The average changes of (L/A ratio) were significant at 24 h after the rapid weight loss, but these changes were not observed at 12 h of recovery time. Statistical analysis of data

TABLE 2 Selected Anthropometric Factors Among Wrestlers in Three Stages of a Weight Loss Program (Mean±SD)

Variables	Pretest	12h	24h	p Value
Body Weight (kg)	67.6±0.7	64.5±0.6*	67.4±0.7	p < 0.05
BMI (kg/m <sup>2</sup> )	22.5±0.21	21.7±0.14*	22.3±0.17	p < 0.05
Body Fat %	6.1±0.18	5±0.19*	5.1±0.12*	p < 0.05
WHR (cm)	0.82±0.08	0.80±0.08*	0.80±0.06	p < 0.05

\*Significant (p < 0.05). Note. Pretest = 1 week before the weight loss program; 12h = 12 hours after weight loss; 24h = 24 hours after weight loss.

showed that the changes in fasting glucose concentration obtained significantly after 12 h of rapid weight loss program, but this pattern was not observed at 24 h of recovery time. The changes in insulin levels were not significant in three stages of weight loss program and shown in Table 5.

Insulin Resistance (HOMA-IR) and Beta Cell Function

The changes in insulin resistance and beta cell function were significant at 12 h recovery from the rapid weight loss program, but these changes were not observed at 24 h of recovery time.

DISCUSSION

Despite the scientific evidence on the deleterious effects of rapid weight loss on health and athletic performance, the application of these methods is still popular among amateur wrestlers. Several studies have been done on the physiological effects of different weight loss patterns and methods. However, in our study before the engaging in main competition we investigated the effect of 1 week of rapid weight loss program on the baseline levels of leptin, adiponectin, leptin-to-adiponectin ratio, insulin sensitivity, and beta cell function of freestyle wrestlers. Wrestlers regularly decrease 4% of their weight through the restriction of calorie (1700±100 kcal) and fluid intake (1100±80CC). This amount of weight loss had a significant reduction on anthropometric parameters (weight, BMI, subcutaneous fat, WHR), but these changes in weight and BMI are not present during the 24 h of recovery time.

TABLE 3 Absolute and Relative Values of Serum Leptin in Three Stage of Rapid Weight Loss Program (Mean±SE)

Variable	Pretest	12h	24h	p Value
Leptin(ng/ml)	7.6±0.31	6.7±0.31	5.6±0.34	p = 0.005*
Leptin/weight	0.10±0.05	0.09±0.05	0.07±0.05	p = 0.004*
Leptin/BMI	0.33±0.01	0.30±0.01	0.24±0.01	p = 0.001*
Leptin/BF%	1.12±0.05	1.33±0.06	1.05±0.06	p = 0.003*
Leptin/WHR	9.26±0.38	8.28±0.36	6.8±0.39	p = 0.007*

\*Significant (p < 0.05). Note. Pretest = 1 week before the weight loss program; 12h = 12 hours after weight loss; 24h = 24 hours after weight loss.

TABLE 4 The Profile of Absolute and Relative Values of Adiponectin Serum and L/A Ratio in Three Stages of Rapid Weight Loss Program (Mean±SE)

Variable	Pretest	12h	24h	p Value
Adiponectin (mu/ml)	7.9±0.37	7.8±0.38	7.8±0.28	p = 0.09
Adiponectin/Weight	0.11±0.6	0.11±0.02	0.15±0.01	p = 0.52
Adiponectin/BMI	0.34±0.01	0.35±0.01	0.34±0.01	p = 0.71
Adiponectin/BF%	1.28±0.07	1.57±0.09*	1.50±0.06	p = 0.006*
Adiponectin/WHR	9.63±0.44	9.48±0.49	9.70±0.32	p = 0.82
L/A Ratio	0.99±0.07	0.88±0.06	0.71±0.05*	p = 0.001

\*Significant (p < 0.05). Note. Pretest = 1 week before the weight loss program; 12h = 12 hours after weight loss; 24h = 24 hours after weight loss.

Rapid Weight Loss and Biochemical Parameters Responses

Several studies have reported conflicting results among athletes. Generally, many factors such as number of samples, baseline body mass index, body fat percentage, duration and type of weight loss program, age, body weight, and physical health statue of subjects can influence the results of different studies (Anderlová et al., 2006; Kotidis et al., 2006; & Yamaner et al., 2010).

In the present study the reduction of 4% weight loss caused a significant reduction in the baseline level of serum leptin. This is in agreement with Pilocova et al. (2003) who used a 5-week weight loss program with an 8 kg weight loss and found a significant decrease in absolute and relative values of leptin levels. In another study Wadden et al. (1998) using a weight loss program for 40 weeks (1200 kcal/daily energy intake), had a 12 kg decrease in body weights and also saw a significant reduction in absolute and relative values of leptin levels.

Murakami et al. (2007) used a 12-week weight loss program with emphasis on caloric restriction (1000–1500 kcal/d) and obtained a 6 kg weight loss, noted a significant reduction in the absolute and relative values of leptin, body fat percentage, and BMI. Sartorio, Agosti, Resnik, and

TABLE 5 The Compared Profiles of Glucose, Insulin, HOMA-IR, and Beta Cell Function Variables in Three Stages of Rapid Weight Loss Program (Mean±SE)

Variable	Pretest	12h	24h	p Value
Glucose (mm/l)	5.32±0.09	4.52±13	5.35±0.09	p = 0.001*
Insulin (µu/ml)	5.12±0.18	4.53±0.18	4.94±0.16	p > 0.05
(HOMA-IR)	1.19±0.05	0.87±0.05	1.17±0.05	p = 0.005*
Beta Cell Function	63.80±2.26	81.33±5.25	61.47±2.16	p = 0.003*

\*Significant (p < 0.05). Note. Pretest = 1 week before the weight loss program; 12h = 12 hours after weight loss; 24h = 24 hours after weight loss.



Lafortuna (2003), reported significant reduction in BMI, body fat percentage, leptin, and insulin after participation in 3 weeks of a weight loss program of caloric restriction, aerobic, and resistance training. Leptin as an anti-obesity hormone is positively correlated with a decrease in visceral and subcutaneous adipose tissue and weight loss program by caloric restriction and increased exercise (more than 800 kcal/d; Hickey et al., 1996).

In the present study, the baseline levels of adiponectin after 12 and 24 h recovery time from weight loss did not result in significant changes, while the pattern of changes in the relative value of adiponectin based on body fat percentage, was significant. Adiponectin as an anti-diabetic and inflammatory hormone is secreted exclusively from adipose tissue and the circulatory concentration is high. Baseline serum levels of this hormone are inversely correlated with body fat percentage, BMI, WHR, and fasting insulin levels, as in the obese, overweight, and diabetics where their baseline levels of adiponectin are reduced. Weight reduction through exercise and caloric restriction has shown significant increases in adiponectin as an anti-atherogenic hormone (Trujillo & Scherer, 2005). Also, in contrast to our findings, Nasseri, Djalali, Keshavarz, and Hosseini (2008) showed a significant increase in absolute and relative values of adiponectin levels following a 5 kg weight loss from a 10-week weight reduction program in 42 healthy women with BMI  $24 \pm 2.4$ ; Gavrilu et al. (2003), Yang et al. (2002), and Esposito et al. (2003), all reported significant increases in baseline levels of adiponectin following weight loss programs.

Generally the results of different studies expressed the sensitivity of adiponectin levels according to body weight changes. In line with this study Varady, Tussing, Bhutani, and Braunschweig (2009) noted no significant changes in baseline concentration of adiponectin by a 5% weight loss through a 3-week weight reduction program. Mäestu et al. (2008) observed no significant increase in adiponectin serum despite the 4% of body weight loss during 5 weeks of restricted calorie intake before the formal competition in male bodybuilders.

Numerous factors may explain these inconsistent results regarding adiponectin levels. These include: the amount of weight loss (more than 5%); source of the reduced fat, visceral versus subcutaneous; the type of diet used; the fitness level of the subjects; increased levels of catecholamines; baseline insulin concentrations; increased baseline levels of IL-6 and TNF- $\alpha$  inflammatory hormones, body mass index; and the baseline levels of testosterone all may have an influence on gene expression and the secretion profile of adiponectin (Abbasi et al., 2006; Borges et al., 2007; Jürimäe et al., 2006; Madsen et al., 2008; Mazzali et al., 2006; Ryan et al., 2003).

#### Leptin-to-Adiponectin Ratio

In this study the leptin-to-adiponectin ratio (L/A ratio) was obtained significantly after the 12 and 24 h of recovery time

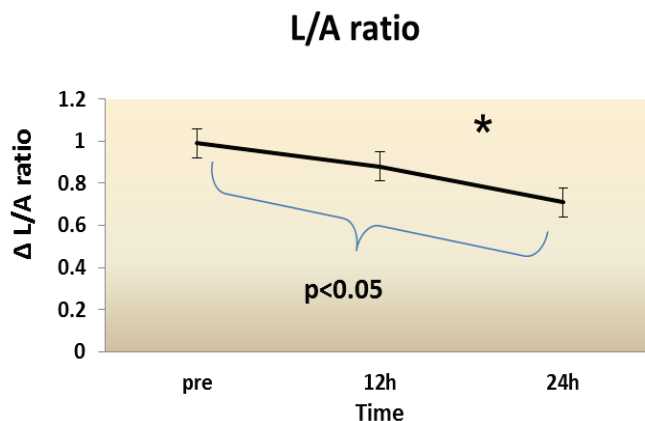


FIGURE 1 Compared profiles of the Leptin/Adiponectin ratio (L/A ratio) in the three stages of the weight loss program (Pretest = 1 week before the weight loss program; 12h = 12 hours after weight loss; 24h = 24 hours after weight loss).

(Figure 1). Leptin and adiponectin are the main secreted hormones from the adipose tissue that as antagonists have paradoxical biological effects with each other. L/A ratio as a valid clinical marker is noteworthy in the study of adipose tissue and diagnosis of the metabolic syndrome, cardiovascular, Type 2 diabetes, and cancer diseases (Finucane et al., 2009). Yadav, Jyoti, Jain, and Bhattacharjee (2011) found a positive correlation between this biochemical marker and anthropometric factors (BMI, BF%, WHR). Smith et al. (2006) showed the L/A ratio is lower in slim and muscular peoples in comparison to obese bodies and body weight is consistent with the variation of the leptin-to-adiponectin ratio. Overall, the results of studies have reported that the L/A ratio is directly related to anthropometric parameters, blood pressure, insulin resistance, and metabolic syndrome. The changes in insulin resistance (HOMA-IR) and beta cell function were significant after 12 h of recovery time (Figure 2). Studies show that insulin resistance is positively correlated with visceral obesity, high body fat percentage, and cardiovascular risk factors. Reduced body fat and regular physical activity are associated with increased insulin sensitivity and beta cell function (Bikman et al., 2008). Wyer et al. (2000) found a significant decrease in insulin resistance and increase in beta cell function after a 5% weight loss over 6 weeks of a weight loss program. Reinehr, Kiess, Kapellen, and Andler (2004) reported a significant reduction in insulin resistance through the loss of a rather small amount (2–3%) of body weight. Mechanisms such as increased metabolism of glucose, fat, sensitivity of receptors in target tissues (muscle, liver), and decreases in pro-inflammatory hormones (IL-6, TNF- $\alpha$ , leptin) and enhanced baseline levels of adiponectin and IL-10 resulted in increased beta cell function and decreased insulin resistance (Soodini & Hamdy, 2004; Wilcox, 2005). The present study utilizing rapid weight loss with an emphasis on reductions in daily caloric intake ( $1700 \pm 100$  kcal) and water ( $1100 \pm 80$ CC) restriction for 1 week prior to engaging in a major tournament, produced significant re-

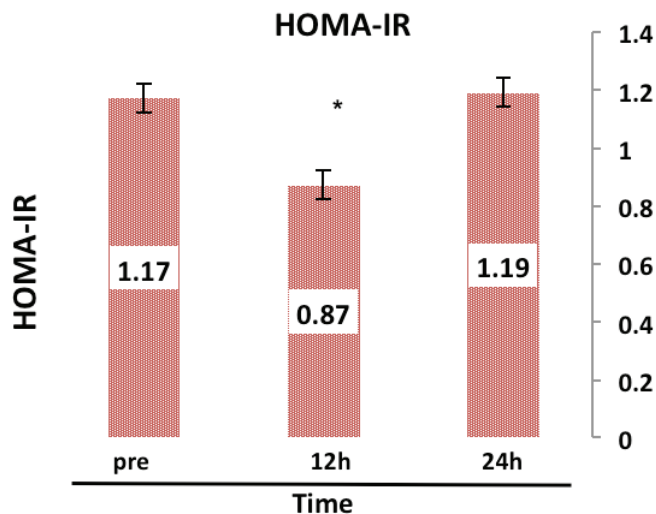


FIGURE 2 Comparing the mean changes of insulin resistance (HOMA-IR) in the three stages of the weight loss program (Pretest = 1 week before the weight loss program; 12h = 12 hours after weight loss; 24h = 24 hours after weight loss).

ductions in selected anthropometric parameters, as well as significant reductions in the baseline levels of leptin, while no significant changes in serum levels of adiponectin were seen.

## CONCLUSIONS

It seems possible that freestyle wrestlers could gradually decrease a greater percentage of their body weight over a longer period of time and take advantage of an increase in baseline adiponectin levels. Decreasing baseline levels of leptin are associated with rapid weight loss in wrestlers. However the role of changes in the levels selected adipocytokines through rapid weight loss have on athletic performance was not investigated after each round of wrestling. Also, it appears that 4% body weight loss had a significant effect on insulin resistance and beta cell function. These findings revealed that despite the negative effects of rapid weight loss on physiological capacity of wrestlers, the combination method of daily caloric and water restriction can reduce the short-term concentrations of pro-inflammatory hormones and increase insulin sensitivity and beta cell function.

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