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The Effects of Whey Protein Supplementation on Performance and Hormonal Adaptations Following Resistance Training in Novice Men

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Authors' Contribution:
A – Study Design
B – Data Collection
C – Statistical Analysis
D – Data Interpretation
E – Manuscript Preparation
F – Literature Search
G – Funds Collection

Key words: protein supplementation, resistance training, hormonal changes

Abstract

Background: *The purpose of this study was to determine the effects of whey protein supplementation on performance and hormonal changes during an 8-week resistance training program in novice weight trained men.*

Material/Methods: *Forty recreational weight trained men were randomly divided into a whey protein supplementation (WP) group (n=20), and a placebo (PL) group (n=20). Each group was given either whey protein or a placebo in a double-blind manner to be taken orally for eight weeks (1.8g/kg/day). WP and PL groups performed the same weight training program 3 days, each week for 8 weeks. The training consisted of 3 sets of 8 repetitions, and the initial weight was 80% of the pre-1RM. Subjects were tested for performance and blood hormone concentrations before and after the 8-week period.*

Results: *The WP group achieved greater increases in body weight, explosive muscular power, muscle strength and blood testosterone when compared to the PL group; however, cortisol concentrations were significantly more reduced in WP group when compared to the PL group.*

Conclusions: *It can, therefore, be concluded that within 8 weeks whey protein supplementation was found to increase explosive muscular power, body weight and muscle strength.*

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Introduction

Resistance training programs are used to achieve many different goals, such as performance improvement, injury rehabilitation, muscle tone improvement and strength improvement [1]. People around the world spend billions of dollars on supplements [2]. On the other hand, improving athletic performance, reducing fatigue, changes in body composition and fitness are the increased requirements for the consumption of nutrient supplements [3]. Protein is one of the most popular dietary supplements marketed to athletes and physically active individuals [4]. Although some studies have demonstrated that protein supplementation in previously untrained adults performing resistance exercise does not provide any benefit in regard to increases in lean body mass or strength [5,6], evidence does support a greater protein need for strength and power athletes compared to endurance athletes and the sedentary population [7,8]. Considering that heavy resistance exercise results in disruption or damage to the active muscle fibers, a greater protein intake may assist in the repair and remodeling process of these fibers [9]. A decrease in muscle damage, attenuation of force decrements, and an enhanced recovery from resistance exercise has been demonstrated in subjects using protein supplements [10,11]. During the manufacturing process of cheese or casein whey proteins are separated from whey liquid (whey) as complete and efficacious proteins with the biological value of 104–159 amino acids, rich in vitamins and minerals required of athletes and active compounds of biological growth factors [12]. To maintain a positive nitrogen balance in strength-trained individuals, it is suggested that they consume protein intake ranging from 1.6 to 1.8 $g \cdot kg^{-1} \cdot day^{-1}$ [13,14]. The combination of resistance training with reduced cortisol concentrations suggests attenuation in the rise of post-exercise muscle degradation [15]. In addition, dietary protein content has also been suggested to influence testosterone concentrations [16] and the hormonal response to a resistance exercise session [17]. Several human [18,19,20, 21] and rodent trials [22,23] demonstrate whey protein's ability to improve body composition (increase in muscle mass and/or a decrease in fat mass) as well as to promote a physiological response that may explain these changes. For example, in a group of healthy adults, in direct comparison to supplementation with casein, whey protein supplementation (30 grams/day) improved body composition (lean mass was maintained, while fat mass was reduced) via enhanced antioxidant (GSH) status [21]. This improvement was obtained without exercise training. Another double-blinded study that used two groups of matched, resistance exercise-trained young men, demonstrated a significantly greater gain in lean body mass and strength in a group provided with whey protein isolate (1.5g/kg/day) compared to a matched group given an equivalent dose of casein [20]. However, there have only been a few studies that have examined the effect of prolonged protein supplementation (e.g. length of a typical off-season resistance training program) on changes in hormonal concentrations in resistance trained men. Studies examining the effect of protein supplementation on strength enhancement are limited and results have been inconclusive. However, other studies did not find any significant changes in the profiles of hormonal responses to resistance exercise due to long-term strength training in adult men [24,25,26,27]. Thus, the purpose of this study was to examine the effect of whey protein supplementation on strength, explosive muscular power, body weight and hormonal adaptations during an 8-week resistance training program in novice weight trained men.

Material and methods

Subjects

Forty healthy recreationally training male students of Guilan University volunteered to participate in this study. Subjects were randomly assigned to either a whey protein supplement group (WP; n =20) or a placebo group (PL; n =20). After signing the informed consent, demographic information regarding each subject was collected. This information included the subject's age, height and body weight (see Table 1). Before undergoing the tests, the subjects were given explanations about the assessment procedures, study objectives, and the possible benefits and risks. The Institutional Review Board of the University approved of the research protocol. According to the medical information questionnaire, all subjects were healthy and none complained of hypertension, a cardiovascular disease, diabetes, lipid disorders, a kidney disease, a liver disease, respiratory and bone injuries and did not report any supplement use in past 6 months. None of them had continuous exercise history. They were recreationally weight trained and did not follow a specific diet. The study protocol was explained to the volunteers, and all of them signed a written consent for the study. Before the study, the subjects were informed about the type, severity and number of days in a week and the time of activities, and they were asked to keep the diet and the intensity of activities constant during the study period, and not to use any other dietary supplements. Their body weight was measured to the nearest 0.1 kg using an electronic body weight scale (Seca 707; Seca GmbH & Co. KG., Hamburg, Germany).

Tab. 1. Study participants' demographics

Parameters	WP group (N=20)	PL group (N=20)
Age (yrs)	21.3 ± 1.2	22.5 ± 3.4
Height (m)	1.74 ± 4.22	1.75 ± 3.52
Body weight (kg)	73.01 ± 5.84	73.42 ± 6.46

Note: Data are presented as means ± standard deviations.
WP: whey protein supplementation group, PL: placebo supplementation group

Experimental design

A double-blind, randomized study was employed using two experimental groups (whey protein and placebo supplementation) who underwent 8 weeks' supplementation. Before starting the training, pre-1 repetition maximum (1RM) values were obtained on the following exercises: leg extension, leg flexion, squat, bench press, lateral pull down, and triceps pushdown. Six different lifts were performed and they were identical to those used in the 1RM measurements. WP and PL groups performed the same weight training program 3 days (Monday, Wednesday, and Friday) each week for 8 weeks. The training consisted of 3 sets of 8 repetitions, and the initial weight was 80% of the pre-1RM. The warm-up prior to each session consisted of 2 sets of 12 repetitions of the first exercise at 40% of the 1RM load, and then 80% of the 1RM load was selected as the load used in testing [18, 28]. When participants were able to perform more than 8 repetitions on the third set, they were instructed to increase their resistance for the next workout. Rest times between sets were 2–3 minutes, and 3–5 minutes elapsed between the 6 different lifts. After selection for either the supplement group or the placebo group, the subjects were required to participate in an 8-week training program, details of which are outlined below. After the 8-week training program, post-testing for 1RM were repeated in the same manner in which they were performed during pre-testing.

Strength measures

Lower and upper body maximal strength was assessed by using 1RM actions. During each testing session subjects performed a 1-repetition maximum (1-RM) strength test for the squat and bench press exercises. The 1 RM tests were conducted as described by Hoffman [29]. Each subject performed a warm-up set using resistance that was approximately 40-60% of his perceived maximum, and then performed three to four subsequent attempts to determine the 1-RM. A 3–5 minute rest period was provided between each lift. No bouncing was permitted, as this would have artificially boosted strength results. Bench press testing was performed in the standard supine position: the subject lowered an Olympic weightlifting bar to midchest and then pressed the weight until his arms were fully extended. The squat exercise required the subject to rest an Olympic weightlifting bar across the trapezius at a self-chosen location. The squat was performed to the parallel position, which was achieved when the greater trochanter of the femur was lowered to the same level as the knee. The subject then lifted the weight until his knees were extended.

Vertical jump

Vertical jump height was measured via a Vertec vertical jump tester (Sports Imports, Hilliard, OH, USA) to give an indication of explosive muscular power [30]. Each subject performed three trials with one minute of rest in between each jump and the highest jump height was used in the data analysis. The following procedure was used for each subject during data collection. The Vertec was adjusted to match the height of the individual subject by having them stand with their dominant side to the base of the testing device. Their dominant hand was raised and the Vertec was adjusted so that their hand was the appropriate distance away from the marker based on markings on the device itself. At that point, subjects performed a countermovement jump. Arm swings were allowed but no preparatory step was performed.

Blood collection and analyses

The subjects were asked to fast for 10 hours before the study. After the first resistance exercise session, blood samples were drawn from an antecubital forearm vein using a 20-gauge needle and Vacutainers to determine serum testosterone and cortisol concentration. For each subject blood samples were obtained, before and after 8 weeks of supplementation (immediately after the first and the last resistance exercise sessions), in the early morning hours and after a 10-h overnight fast in order to minimize the effects of diurnal hormonal variations. The blood was processed and centrifuged, and the resultant serum was stored at −80°C until analyzed. Serum total testosterone and cortisol were determined in duplicate by using standard RIA procedures and were assayed via ELISA kits obtained from Diagnostic Systems Laboratories (Webster, TX. OH).

Supplement schedule

The whey protein supplement (ultimate nutrition, Farmington Inc, CT, USA) and placebo (the placebo content of the supplement consisted of starch) was in powder form and provided in individual packets. The only permitted supplement was pure whey protein powder containing no additional ingredients. Daily whey protein supplementation was 1.8g/kg/day parceled into three equal dosages to be consumed with each major meal. The contents of each packet were mixed with 500 ml of water. Subjects consumed one drink every morning, the second daily drink following their exercise session and the third daily drink in the evening. On non-training days, the WP and PL groups ingested 1 dose of the WP or PL supplement in the morning and once again in the evening. The subjects consumed the supplements for 8-week.

Statistical analyses

Statistical analyses were performed using the Statistical Package for Social Sciences (SPSS) for Windows software (version 16.0; SPSS Inc.). Descriptive statistics were calculated as the mean and standard deviations (Mean ± SD). Changes from baseline were assessed using the paired sample t–test. In addition, PRE – POST comparisons between groups in performance measures were analyzed with independent student's t-tests. The level of significance for this investigation was set at $P<0.05$.

Results

Performance

Significant increases in strength from PRE occurred for both WP and PL in the 1-RM squat and 1-RM bench press (see Table 2). However, strength comparisons showed that subjects in WP had significantly greater improvement in 1-RM squat and 1-RM bench press strength compared to the PL group.

The mean changes in explosive muscular power performance in WP and PL groups are shown in Table 2. Significant increases in power from PRE occurred for both WP and PL in the vertical jump. However, power comparisons showed that subjects in WP had significantly greater improvement in explosive muscular power performance compared to the PL group.

Hormonal adaptations

A significant increase in blood testosterone from PRE occurred for both WP and PL groups (see Table 2). However, blood testosterone comparisons showed that subjects in WP had significantly greater increase in blood testosterone compared to the PL group.

A significant decrease in blood cortisol from PRE occurred for both WP and PL groups (see Table 2). However, blood cortisol comparisons showed that subjects in WP had significantly greater decrease in blood cortisol compared to PL group.

In between groups, a significant difference was observed in entire six factors at post-test averages, but not in pre-tests.

Body weight

8 weeks of whey protein supplementation resulted in a significant body weight increase from PRE in both WP and PL groups. However, body weight comparisons showed that subjects in WP had a significantly greater increase in weight (see Table 2).

Tab. 2. Measures of body weight, strength and hormonal adaptation in the WP (N=20) and PL (N=20) groups during the pre and post-supplementation period. Data presented as mean ± SEM

	Pre	Post	Pre	Post
	WP		**PL**	
Upper body Strength	80 ± 7.07	87.12 ± 6.35***	83.12 ± 6.53	86.75 ± 6.54*
Lower body Strength	88.87 ± 5.87	94.62 ± 6.45***	88.37 ± 7.87	92.12 ± 8.59*
Body weight (kg)	73.01 ± 5.84	75.41 ± 6.12***	73.42 ± 6.46	73.87 ± 6.53*
Vertical Jump (cm)	64.11 ± 4.12	69.42 ± 5.14***	66.31 ± 6.44	68.62 ± 7.33*
Cortisol (mg %)	19.13 ± 2.06	17.77 ± 2.48***	19.48 ± 2.08	19.13 ± 2.03
Testosterone (ng/ml)	5.95 ± 0.32	6.72 ± 0.24***	6.02 ± 0.42	6.44 ± 0.41

WP: whey protein group, PL: placebo group.

* Significantly different from corresponding to pre training value; ** Significantly different between WP and PL

Discussion

Resistance training has been shown to increase strength and body weight [31,32]. Resistance training alone stimulates muscle protein metabolism, which can lead to muscle growth and strength increase [9, 33, 34]. To stimulate muscle protein synthesis, however, amino acid availability is important, especially in the first few hours after exercise [35]. Protein supplementation increases muscle protein synthesis without a corresponding increase in protein breakdown, which results in a net positive protein balance, thus allowing for maximal recovery, hypertrophy, and strength gains [35]. “Fast” proteins, such as whey, are characterized by the rapid appearance of their amino acid constituents in the blood and have been shown to elicit strength gains and improve muscle protein balance [34]. The results of this study demonstrated that even though both groups demonstrated significant strength increases over time, the whey protein supplemented (WP) group showed greater improvements in strength when compared to the placebo (PL) group. These findings were similar to the study by Bird et al [36]. This may be attributed to a higher average weekly training volume and intensity seen during the 8-week study for the squat and bench press exercise in WP compared to PL. Interestingly, Kraemer et al. [37] reported no differences in the training volume or intensity in experienced resistance-trained men during several days of protein supplementation. However, they suggested that supplementation for a longer period of time may have resulted in more favorable outcomes. It is thought that protein supplementation can stimulate muscle protein synthesis to counteract the deleterious effects of muscle degradation seen after bouts of resistance exercise [34]. If protein degradation is reduced with a concomitant increase in protein accretion, the resulting effect would generate a greater stimulus for muscle growth and enhanced recovery, potentially resulting in greater strength gains [10,11]. Still, some investigators have shown augmented strength gains from protein supplementation, for example, Coburn et al. [31] randomly assigned adult male subjects to a supplement (20 g whey protein, 6.2 g leucine), carbohydrate placebo (26.2 g maltodextrin), or control group for 8 weeks of unilateral (nondominant limb) leg extension resistance training. Even though the supplement contained similar quantities of whey protein and leucine as the current study, the protein supplemented subjects demonstrated a 30% increase in strength in the trained limb which was significantly higher than the strength increase (22%) achieved by the carbohydrate placebo group. No strength changes were observed in the control group. Similarly, Willoughby et al. [32] compared the effects of a 10 week resistance training program combined with 20 g protein (14 g whey and casein protein, 6 g free (essential and non-essential) amino acids) or 20 g dextrose placebo ingested 1hr before and after exercise on muscular strength in untrained males. Others have reported no effect [38,39].

The vertical jump test is a simple and reliable test that can provide useful information about explosive muscular power and performance characteristics of athletes [30]. Significant changes were seen during the 8-week resistance training program in any of the power performance measures for either group. Although WP has been shown to significantly enhance power performance, these findings were similar to the study by Andersen et al [40] and the study by Buckley et al. [41]. Other studies have shown no significant differences between subjects consuming a protein supplement compared to a placebo [42]. However, neither of those studies used experienced strength/power athletes. It is likely that the lack of specificity between the training program and exercises used to assess power performance in this study was the primary factor that negated any potential effects of the supplement on power assessments.

8 weeks of whey protein supplementation resulted in a significant increase in body weight. However, body weight comparisons showed that subjects in WP had significantly greater increase weight. These findings were similar to the some other studies [18,19,20,21,22,23], although a number of resistance training studies [43] involving supplementation have reported no significant changes in body weight during a 12-week protein supplementation period in experienced resistance trained athletes. Although some studies have demonstrated that protein supplementation in previously untrained adults performing resistance exercise does not provide any benefit in regard to increases in lean body mass [5,6].

Blood concentrations of testosterone stimulate muscle protein accretion [44]. Testosterone also increases protein synthesis by binding to the androgen receptor for the complex to become a transcription factor and thirdly by possibly activating muscle satellite cells, which is important because gene transcription is an initial target for the modulation of protein synthesis [45,46]. Resistance training is associated with significant elevations in anabolic hormones such as testosterone [47]. The results of this study demonstrated a significant increase in blood testosterone from PRE in both WP and PL groups. However, blood testosterone comparisons showed that subjects in WP had a significantly greater increase in blood testosterone compared to the PL group. These findings were similar to the study by Kraemer et al. [36] who reported that untrained men are able to develop an exercise-induced increase in testosterone. Thus increases in anabolic hormones such as testosterone help to enhance protein synthesis in skeletal muscle and to promote muscle growth [48].

Cortisol is an adreno-cortical steroid hormone released into the body from the adrenal cortex in response to stressful physical or psychological stimuli [49]. Resistance training may also have led to an overall reduction [44,50] or similar [24,51] cortisol responses to exercise loading in men. Although the results of this study demonstrated a significant decrease in blood cortisol after supplementation. This improved hormonal response may help to reduce the significant decrease in muscle glycogen that occurs during exercise, and result in an enhanced anabolic environment where muscular adaptations and recovery can occur. Furthermore, protein supplementation increases the availability of amino acids which may result in an increased uptake of amino acids by the muscle. Increased uptake of amino acids by the muscle enhances net muscle protein balance and improves the anabolic environment [52].

Comparison of our results with other studies is difficult because the magnitude of exercise induced loss in maximal strength or muscle power has not been reported systematically. Also, for most healthy people, a high-protein diet generally is not harmful if followed for a short time, such as three to four months. However, the risks of using a high-protein diet for a long time are still being studied. Several health problems may result if a high-protein diet is followed for an extended period, such as a metabolic disease, a cardiovascular disease, renal dysfunction, bone disorders and a liver disease [53].

Conclusion

In conclusion, the results of this investigation suggested that 8-week resistance training combined with the timed ingestion of whey protein supplementation increase explosive muscular power, body weight and muscle strength (upper and lower body strength) in novice weight trained men. Additionally, the strategic consumption of a daily whey protein supplementation (1.8g/kg/day) parceled into three equal dosages represents a simple but effective strategy that enhances performance during resistance training.

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