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Effects of Meal Form and Composition on Plasma Testosterone, Cortisol, and Insulin Following Resistance Exercise

Richard J. Bloomer, Gary A. Sforzo, and Betsy A. Keller

The purpose of this study was to examine the effects of postexercise feeding on plasma levels of insulin, testosterone, cortisol, and testosterone:cortisol (T:C). Ten experienced, resistance trained males (20.7 ± 0.95 years) were given whole food (WF: protein 38 g; carbohydrate 70 g; fat 7 g), a supplemental drink (SD; isocaloric and isonitrogenous to WF), an isocaloric carbohydrate beverage (C), or a placebo beverage (P) immediately, 2 and 4 hours after a standardized weight training protocol on 4 days, each separated by 1 week, in a repeated measures design. Subjects also received a standardized meal at 7 and 12 hours postexercise. Insulin, testosterone, and cortisol were measured pre-exercise and during 24 hours of recovery (at 0.5, 2.5, 4.5, 8, and 24 hours) using venous blood samples. Significant (condition × time) interactions were found for insulin, testosterone, and T:C, but not for cortisol ($p < .05$). The SD yielded a greater response for insulin than all other conditions. Conversely, P demonstrated the greatest values for testosterone and T:C at 2.5 and 4.5 hours postexercise. Cortisol did not vary between conditions and there were no condition effects for insulin, testosterone, cortisol, and T:C at 8 or 24 hours. In conclusion, the efficacy of postexercise feeding for optimizing T:C and muscle growth is unclear; however, consumption of SD appears to maximize circulating insulin for several hours following resistance exercise.

Key Words: hormones, dietary supplements, muscle hypertrophy, nutrition support, exercise recovery

Introduction

During and after resistance exercise (RE), optimizing the hormonal environment toward anabolism may be a key factor in promoting muscle hypertrophy and strength gain. Throughout exercise, circulating levels of both anabolic (e.g., testosterone) and catabolic (e.g., cortisol) hormones increase with intensity (11). While testosterone is known to promote hypertrophy (12), increases in corticosteroids after exercise oppose new tissue remodeling and accelerate protein degradation for up to 64 hours (6).

At the time the research was conducted, the authors were with the Department of Exercise and Sport Sciences at Ithaca College, Ithaca, NY 14850. R.J. Bloomer is now with the University of North Carolina at Greensboro.

Due to these effects, athletes sometimes use androgens to reduce catabolic influences and promote anabolism. In addition, the hormone insulin appears to antagonize the negative effects of cortisol on both synthesis and degradation pathways of protein turnover (2). Insulin's efficacy as an anabolic agent has previously been observed in both healthy (2, 10) and diseased or injured populations (9) investigated under intravenous administration.

Dietary manipulation (e.g., increasing caloric intake or varying macronutrient composition of the diet) can substantially impact plasma insulin concentration. Variation in the insulin response is observed when comparing macronutrient form (8), glycemic index (18), and particle size (13). Further, dietary intake may influence resting levels of testosterone (26). Thus, specific alterations to the postexercise meal(s) could potentially affect the hormonal environment to favor muscle growth.

For this reason, the use of postexercise dietary supplements to promote anabolism (4, 14, 26), decrease markers of muscle damage (3), and increase glycogen resynthesis (28) is quite popular. Indeed, feeding following exercise may prove beneficial, because protein synthesis is elevated postexercise (25), particularly under conditions of acute hyperaminoacidemia (1, 25).

Ingestion of a liquid carbohydrate and protein supplement after exercise may elevate insulin (4) and testosterone, and attenuate cortisol. While data appear to support the insulin elevating effects of postexercise feeding, the effects on testosterone and cortisol concentrations are unclear. The aim of this study was to compare the effects of three meals (varying in energy and macronutrient composition) on insulin, testosterone, and cortisol concentrations during 24 hours of exercise recovery.

Methods

Subjects

Subjects were 10 healthy, currently training males, aged 20 to 25 years, with more than 2 years of RE experience. None of the subjects had used anabolic agents in the past. Drug-free status and training history were determined via a questionnaire, and all subjects provided informed consent according to guidelines of the Institutional Review Board for Human Subjects Research. Table 1 presents descriptive characteristics of the subjects.

Experimental Design

During four training sessions, separated by at least 1 week, subjects performed a RE protocol followed immediately (within 15 min) by consumption of either (a) a whole food meal (WF; baked, boneless/skinless chicken breast meat and boiled, long grain white rice: protein 38 g; carbohydrate 70 g; fat 7 g), (b) a mixed-nutrient supplemental drink (SD; Myoplex Mass, Experimental and Applied Sciences, Golden, CO—blend of whey, casein, and milk protein; maltodextrin, sucrose, and dextrose; canola, medium-chain triglyceride, and borage oil; isocaloric and isonitrogenous to the whole food meal), (c) a carbohydrate beverage (C; maltodextrin and fructose; isocaloric to whole food meal and supplemental drink), or (d) a placebo beverage (P; carbohydrate 3 g). In all conditions, the same meal was given at 2 and 4 hours postexercise, and subjects consumed 500 ml of water during each meal. The order of conditions was counterbalanced among the subjects in double blind

Table 1 Subject Characteristics and 1 RM (N = 10)

Variable	Mean	*SD*
Age (years)	20.70	0.95
Years training	5.40	2.46
Weight (kg)	93.08	19.15
Height (cm)	181.43	8.38
Body fat %[a]	17.28	6.25
Fat free mass (kg)	76.10	10.54
Fat mass (kg)	16.97	9.46
Squat (kg)	137.7	26.40
Bench press (kg)	109.6	24.91
Barbell row (kg)	85.7	12.31
Military press (kg)	75.7	16.77
Lat pulldown (kg)	86.8	13.59
Close grip bench press (kg)	97.1	19.93

[a]Seven site skinfold formula (17).

fashion. Under all conditions, every subject consumed a standardized whole food meal (described above) at 7 hours postexercise. They were also given a powder mix of identical caloric and macronutrient composition to the supplemental drink, to consume with water at 12 hours postexercise. No other food was consumed during the 24-hour study period. Blood samples were taken pre-exercise and five times postexercise (at 0.5, 2.5, 4.5, 8, and 24 hours) and were subsequently analyzed for insulin, testosterone, and cortisol.

On the first day of testing, all subjects were given water ad libitum during exercise. Hydration level is known to affect hormonal events and provides an important signal for cellular direction of protein turnover (27). Due to this fact, water intake on the subsequent three visits was matched to the volume consumed during exercise on the first day. Subjects were also required to consume similar meals prior to each testing day, with the last meal consumed at least 8 hours prior to testing. Food intake records were maintained for the 2 days preceding each experimental day to assess dietary adherence. Diet analyses (Dine Systems, Amherst, NY) confirmed that these dietary guidelines were followed throughout the course of the study.

Resistance Exercise Protocol

Subjects visited the lab 1 week prior to the first experimental trial to practice the RE protocol and to determine their one repetition maximum (1RM) for each exercise.

The RE protocol, in order, included the barbell squat, barbell bench press, barbell row, barbell military press, latissimus dorsi pulldown, and barbell close-grip bench press. On each testing day, subjects performed a standard warm-up, and all exercises were done for three sets at 75% 1RM. Each set was performed to a point of momentary muscular failure with 120-s rest between sets. The exercise session was identical for a given subject on all four visits.

Blood Sampling and Analysis

On each of the 4 test days, subjects reported to the laboratory and rested quietly for 20 min. After this time, 8 hour post-absorptive, baseline blood samples were taken from the antecubital vein between 0700–0800 hours and again at 0.5, 2.5, 4.5, 8, and 24 hours postexercise. Samples were allowed to clot at room temperature for 30 min and serum was then separated by centrifugation for 10 min at 1,500 rpm. The sampling procedure was consistent throughout the experiment. Aliquots for testosterone analyses were refrigerated at 2 °C and assayed within 24 hours using the Ciba Corning ACS:180™ competitive chemiluminescent immunoassay procedure (SmithKline Beecham Clinical Laboratories, Syracuse, NY). Aliquots for cortisol and insulin were frozen at –20 °C with cortisol determined by fluorescence polarization and insulin measured by standard radioimmunoassay (Radioimmunoassay Core Laboratory, Washington University School of Medicine, St. Louis, MO). All samples were assayed in duplicate. Intra- and interassay variability was less than 5%. Hematocrit and hemoglobin values were also determined in duplicate using centrifugation and the Cyanmethemoglobin Procedure (24), respectively, prior to performing analyses to adjust hormone concentrations for changes in plasma volume (7).

Statistical Analysis

Two-way repeated measures ANOVAs for Conditions (4) and Times (6) were done for insulin, testosterone, cortisol, and T:C. Where appropriate, significant interactions and main effects were further evaluated with analyses of simple main effects and Tukey HSD. The level of significance for this investigation was set at $p < .05$.

Results

Insulin

Placebo concentrations were lower than any meal values at 0.5, 2.5, and 4.5 hours into recovery ($p < .05$). Supplemental drink insulin concentrations were higher than the other meals at 0.5 hour and also greater than WF and P through 4.5 hours postexercise (Figure 1). Insulin was lowest across all conditions at baseline and at 24 hours postexercise, in the fasted state. Insulin levels were near baseline at 8 hours postexercise, and no significant differences existed between conditions at 8 or 24 hours.

Testosterone

Figure 2 shows that testosterone levels were greatest at baseline and 24 hours postexercise, yet there were no significant differences between conditions at either time. Testosterone levels generally decreased postexercise and were greater in the P condition through 4.5 hours postexercise than in any other condition.

Cortisol

As with testosterone, the highest plasma cortisol concentrations occurred at baseline and 24 hours postexercise with no differences between conditions (Figure 3). A significant main effect for time demonstrated that cortisol decreased greatly at 2.5

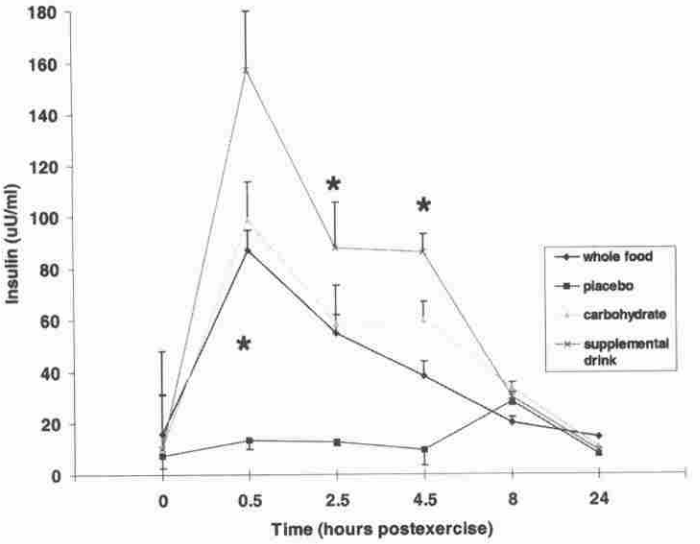

Figure 1 — Postexercise insulin (Mean ± *SEM*) for each condition (*N* = 10, except 24 hours whole food, *N* = 9). *0.5 hour: placebo < all other conditions; supplemental drink > carbohydrate and whole food ($p < .05$). *2.5 hours: placebo < all other conditions; supplemental drink > whole food ($p < .05$). *4.5 hours: carbohydrate and supplemental drink > placebo; supplemental drink > whole food ($p < .05$).

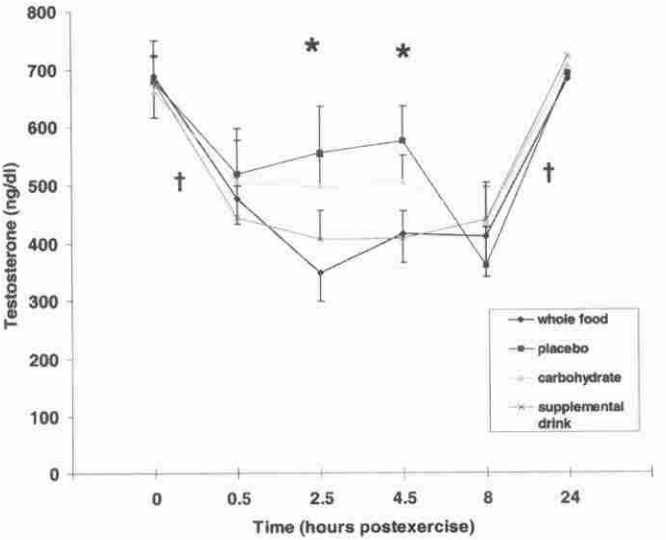

Figure 2 — Postexercise testosterone (Mean ± *SEM*) for each condition (*N* = 10, except 24 hours whole food, *N* = 9). *2.5 hours: placebo > whole food and supplemental drink; whole food < carbohydrate ($p < .05$). *4.5 hours: placebo > whole food and supplemental drink ($p < .05$). †Pre-exercise and 24 hours differed from 0.5, 2.5, 4.5, and 8 hours ($p < .05$).

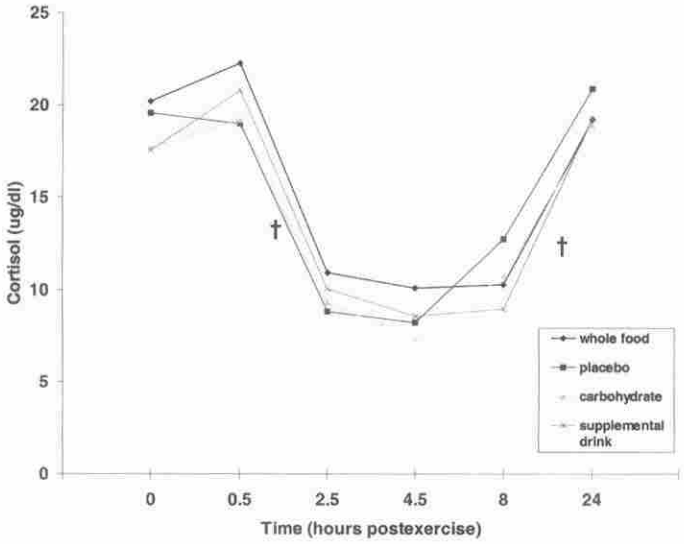

Figure 3 — Postexercise cortisol (Mean ± *SEM*) for each condition ($N = 10$, except at 24 hours whole food $N = 9$). †Pre-exercise, 0.5, and 24 hours > 2.5, 4.5, and 8 hours ($p < .05$).

hours postexercise, remained stable across all conditions until 8 hours postexercise, then returned to baseline values by 24 hours postexercise.

Testosterone:Cortisol

T:C largely reflects the drastic change in cortisol over time; however, Figure 4 illustrates a significant interaction at 2.5 and 4.5 hours postexercise. T:C in both the P and C conditions was significantly greater than the WF at 2.5 hours, and greater than both the WF and SD at 4.5 hours. By 24 hours postexercise, T:C had returned to near baseline values across all conditions.

Discussion

The main finding of this study was that a mixed-nutrient supplemental drink, consumed after RE elevated plasma insulin more at 0.5 hour into recovery than an isocaloric, isonitrogenous whole food meal, carbohydrate beverage, or placebo beverage. These data corroborate previous studies that showed greater elevations in insulin with a mixed-nutrient feeding compared to a carbohydrate only feeding (28). Only this investigation and that of Chandler et al. (4) have reported greater elevations in plasma insulin with mixed feedings compared to solely carbohydrate feedings under isoenergetic conditions. Conversely, other work showed similar (22) and greater elevations (23) in insulin response with isoenergetic carbohydrate–only feedings compared to mixed carbohydrate-protein feedings.

These differing results may be caused by a higher glycemic index and lower molecular weight of the meal, and to differing protein sources (i.e., whey protein in liquid feeding) between meals (13, 18). These disparities may result in greater plasma accumulation of insulin with the liquid, mixed-nutrient feeding, possibly

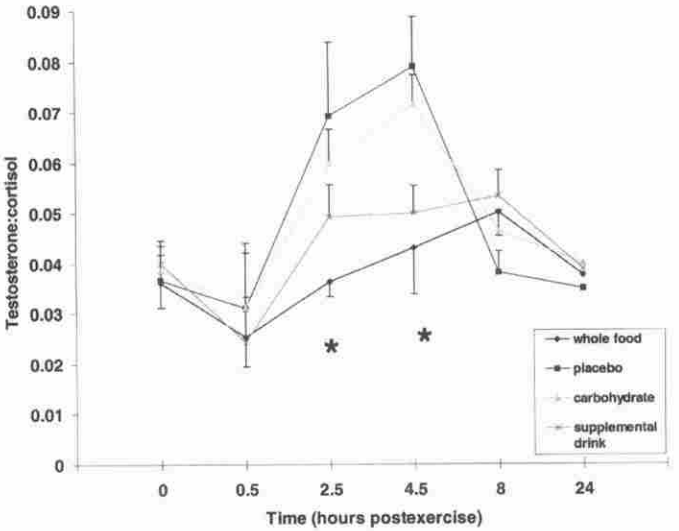

Figure 4 — Postexercise testosterone:cortisol (Mean ±*SEM*) for each condition ($N = 10$, except at 24 hours whole food $N = 9$). *2.5 hours: placebo and carbohydrate > whole food ($p < .05$). *4.5 hours: placebo > whole food and supplemental drink; carbohydrate > whole food and supplemental drink ($p < .05$).

because of faster gastric emptying, digestion, absorption, and assimilation rates (5, 8). This would be equally true when comparing the mixed-nutrient liquid and solid food feedings. Also noteworthy, from a practical standpoint, our subjects reported it was easier during the immediate postexercise period to consume a liquid feeding (i.e., palatability, convenience, time) than a whole food meal. This is important as individuals may neglect adequate caloric intake immediately postexercise if they find food consumption difficult, particularly for athletes involved in multiple daily exercise sessions.

Insulin promotes both anabolic and anticatabolic activity in muscle by stimulating protein synthesis (10), decreasing proteolysis (2) possibly via cortisol suppression (15), and enhancing nitrogen retention (19). Previous studies (1, 25) have reported that protein synthesis is enhanced following exercise, and is further stimulated by amino acid ingestion. Therefore, in the present study, although carbohydrate-only ingestion increased postexercise insulin, protein synthesis and tissue growth are projected to be relatively less with C than with SD due to a lack of supplemental amino acids.

Moreover, based on previous data, the carbohydrate-protein mixture would be predicted to cause the most efficient glycogen resynthesis following exercise, as greater insulin availability promotes muscle uptake of glucose and amino acids (28); however, this was not determined in this investigation. Based solely on the insulin data, the SD created the most favorable postexercise environment for anabolic activity; however, testosterone and cortisol results also merit consideration.

Testosterone was generally depressed across all conditions for 8 hours postexercise and returned to near baseline values at 24 hours. The magnitude of the testosterone decrease was least in P, suggesting that plasma testosterone is highest while fasting. These results are in agreement with Chandler et al. (4) who reported a

decrease in testosterone following feeding, with the smallest decrement observed in the control condition. Kraemer et al. (14) observed a similar phenomenon following consumption of a carbohydrate-protein supplement compared with a noncaloric placebo. Data from the present study display an inverse relationship between testosterone and insulin, also shown in previous work (4, 14). Specifically, when insulin levels were lowest, testosterone levels peaked, and when insulin levels were highest, testosterone levels reached a nadir. Mechanistically, insulin infusion can increase the clearance rate of androgenic precursors (DHEA and DHEA sulfate) but not testosterone (16). However, it has been shown that insulin is negatively correlated with testosterone (21). In contrast, other data have shown testosterone increased with acute and chronic elevations in insulin (20). Taken together, these findings fail to explain the results obtained in the present study. Additional research is needed to more fully elucidate how a meal might attenuate the typical exercise-induced rise in testosterone.

Cortisol is expected to increase in an exercise intensity-duration dependent manner (11). In this study, cortisol rose (nonsignificantly) initially after exercise before dropping significantly below baseline values at 2.5 hours in all feeding conditions. Diminished cortisol was still evident at 8 hours postexercise but returned to pre-exercise levels at 24 hours postexercise. This pattern of change in cortisol mirrors the documented circadian rhythm of this hormone (12), suggesting that varying the composition or form of postexercise feeding had little effect on circulating cortisol levels. Although insulin was greatest in the liquid, mixed-nutrient condition, it did not appear to further reduce plasma cortisol levels as might be expected from previous research (15). Without direct observations of protein turnover, it is difficult to know whether the mixed-nutrient SD or the P condition is more effective in promoting an acute state of postexercise muscle anabolism.

In summary, there appear to be few beneficial effects of varying postexercise meal form or composition on regulating plasma testosterone and cortisol. These hormones appear to be regulated more by diurnal variations than acute feeding patterns. Although T:C was highest in the fasted state, the SD promoted the greatest increase in insulin during the immediate 4–5 hours postexercise. The efficacy of a particular postexercise feeding for optimizing T:C and muscle tissue growth is unclear. However, a mixed-nutrient SD will promote postexercise hyperinsulinemia which may mediate muscle tissue growth and enhance glycogen resynthesis.

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This experiment conforms with the policies of the U.S. Department of Health, Education, and Welfare and the American Physiological Society. Our protocols were approved by our Institutional Review Board on Human Subjects Research.