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Casein Protein Supplementation in Trained Men and Women: Morning versus Evening

JOSE ANTONIO‡, ANYA ELLERBROEK*, COREY PEACOCK‡, and TOBIN SILVER‡

Department of Health and Human Performance, Nova Southeastern University, Davie, FL, USA

*Denotes undergraduate, ‡Denotes professional author

ABSTRACT

International Journal of Exercise Science 10(3): 479-486, 2017. The purpose of this investigation was to determine the effects of casein supplementation (54 grams) in the morning (Casein-MOR) or evening (Casein-EVE) (90 minutes or less prior to sleep) on measures of body composition and exercise performance in trained men and women. Twenty-six healthy men and women who had been training regularly for greater than 12 months completed this 8-week study (mean±SD; Casein-MOR group [n=14, seven male, seven female]: 30.0±8.2 yr; 170.7±9.5 cm; 70.9±13.9 kg. Casein-EVE group [n=12, nine male, three female]: 28.9±9.5 yr; 172.9±7.3 cm; 72.6±10.9 kg). Subjects in each group supplemented with casein protein (54 grams) either in the morning (prior to 12:00pm) or evening (~90 minutes or less prior to sleep). Subjects were advised to not significantly alter their training program as well as to keep a diary of their workouts. Body composition was assessed via the Bod Pod®. In addition, subjects provided dietary self-reports via MyFitnessPal®. Approximately 24 daily dietary self-reports were provided from each subject that self-monitored their diet. The investigators monitored their diet throughout the study. Both the Casein-MOR and Casein-EVE groups consumed significantly more protein (post versus pre; $p<0.05$); however, there were no between-group differences regarding protein intake. Furthermore, there were no within- or between-group differences for any other measure. In trained subjects who did not significantly alter their training program, the addition of 54 grams of casein protein in the morning or evening had no significant effects on body composition. Furthermore, the additional consumption of protein calories did not result in an increase in fat mass despite the fact that exercise volume did not change.

KEY WORDS: Sports nutrition, supplements, overfeeding, nutrient timing

INTRODUCTION

Previous investigations have shown that dietary amino acids are absorbed into the bloodstream from the gut at varying rates(5). It has been shown for instance that whey and casein protein have different absorption characteristics. Whey protein has been shown to produce a very quick rise in plasma amino acids and then decreases to baseline levels in approximately 3 hours (i.e., it is a 'fast' protein). On the other hand, casein protein produces a sustained increase in plasma amino acids of ~7 hours (i.e., a 'slow protein'); however, the initial spike is much less dramatic when compared to whey (4). In fact, when looking at measures of net leucine balance, casein may be superior to whey (4, 5). Conversely, other investigators have found that consuming whey protein induces greater increases in muscle protein synthesis in comparison to micellar casein both at rest and after resistance exercise in healthy elderly men (7). It has been posited that one can take advantage of casein's 'slow' properties via the ingestion of it prior to nighttime sleep. However, there is very little data to show if the 'slow' properties of casein can produce an anabolic effect during the night when ingested prior to sleep. Res et al. was the first study to show that protein ingested immediately before sleep is effectively digested and absorbed; subsequently, muscle protein synthesis and whole-body protein balance is enhanced post-exercise overnight (15). Currently, there is little research on the effects of protein timing as it relates to the nighttime feeding of casein (10-13, 15, 16). However, it is not known if chronic feeding of casein protein immediately pre-sleep would have anabolic effects in the long run. Therefore, the purpose of this investigation was to compare the effects of morning versus evening supplementation of casein protein on body composition in exercise-trained men and women over an 8-week treatment period.

METHODS

Participants

Twenty-six exercise-trained men and women completed this 8-week study. Eighteen of the subjects had been performing resistance training on average for greater than one year on average > 3 sessions per week. The remaining 8 subjects had extensive training (>5 years experience) aerobically (i.e., stand up paddling). Originally there were 35 subjects that volunteered. Five subjects dropped out with no reason given. Three were dropped from the study due to lack of compliance with protein consumption. And one dropped out due to an illness that prohibited the subject from training. Subjects were randomly assigned to a group that consumed casein protein either in the morning or evening (Casein-MOR versus Casein-EVE). They were instructed to consume casein protein in addition to their regular diet. Subjects were instructed to maintain their usual exercise training habits. Nova Southeastern University's Human Subjects Institutional Review Board in accordance with the Helsinki Declaration approved all procedures involving human subjects and written informed consent was obtained prior to participation.

Protocol

All subjects were instructed to keep food and fluid intake diaries, which were captured by use of integrative mobile technology (MyFitnessPal®). Subjects kept a diary three times per week of their food intake and were monitored regularly by the investigators to insure compliance. Thus on average, each subject provided 24 daily food logs. This included two weekdays and one weekend day. The use of a mobile app for quantifying food intake has been previously shown to be an effective monitoring tool (17).

Casein protein (MusclePharm®) was provided at no cost to the research subjects. Subjects were instructed to consume two scoops of protein powder equaling 54 grams of casein either in the morning (before 12:00 noon) or evening (90 minutes prior to going to bed). The 54 gram dose was chosen because 1) it would substantially increase the total daily protein intake and 2) it represented the dose provided by the manufacturer (based on two scoops) as found on the supplement facts panel. Thus, it was a convenient and easy to measure dose for each subject. It should be noted that unlike the pre-sleep time frame (90 minutes prior to going to bed), the morning time frame was much broader to accommodate the widely varying schedules of our study subjects. That is, some subjects would arise at 6am and consume their protein supplement within the hour. On the other hand, other subjects might arise a few hours later (e.g., 10:00am). Ultimately, the goal was to have our two groups protein-equated. That is, in order to determine the effects of nighttime feeding, it would be judicious to have another group consume protein at a time that was substantially different from nighttime.

Height was measured using standard anthropometry and total body weight was measured using a calibrated scale. Body composition was assessed by whole body densitometry using air displacement via the Bod Pod® (COSMED USA, Concord CA). All testing was performed in accordance with the manufacturer's instructions. Subjects were tested while wearing only tight fitting clothing (swimsuit or undergarments) and an acrylic swim cap. The subjects wore the same clothing for all testing. Thoracic gas volume was estimated for all subjects using a predictive equation integral to the Bod Pod® software. The calculated value for body density used the Siri equation to estimate body composition. Data from the Bod Pod® included body weight, % body fat, fat free mass and fat mass. Measurements were taken with each subject at approximately the same time of day pre and post.

Performance testing included the one repetition maximum (1-RM) bench press and repetitions to failure (RTF) at 60% of the bench press 1-RM. Performance tests were conducted by certified strength and conditioning specialists. All subjects were familiar with the performance tests prior to entering the laboratory. In general, each subject performed a movement specific warm up prior to the test (i.e., 3 sets on the bench press at progressively higher submaximal loads). Subjects then rested for 2-3 minutes prior to commencing the 1-RM bench press. A maximum of five attempts was allowed for the 1-RM bench press. Once the subject achieved their 1-RM, they rested for 3-5 minutes prior to commencing the RTF at 60% of the 1-RM bench press. The maximal number of repetitions was subsequently determined. In addition, a subset of subjects was tested on a stand-up paddling (SUP) ergometer (KayakPro USA LLC, Miami Beach FL). After performing the standard calibration as directed by the manufacturer, subjects completed 5-10 minute warm up on the SUP ergometer and then completed a 500-meter time trial.

Each subject followed their own strength and conditioning program. The investigators were in regular contact with each subject to ensure that each subject completed a training log. The volume load (i.e., total weight lifted per week) was determined for each treatment period.

Statistical Analysis

A 2-way analysis of variance (ANOVA) was used to analyze the data with a $p < 0.05$ considered significant. We compared pre versus post (within groups) as well as between group data. Data are expressed as the mean±SD. The statistical analysis was completed using Prism 6 GraphPad Software (La Jolla California).

RESULTS

There was a significant increase it absolute and relative protein intake pre versus post in both groups with no between group differences. However, there were no significant differences in body composition or exercise performance within or between groups (Tables 1-3) for any of the parameters measured.

Table 1. Body composition.

	Casein-MOR		Casein-EVE	
	Pre	**Post**	**Pre**	**Post**
Weight kg	70.9±13.9	71.2±13.1	72.6±10.9	73.7±9.9
Fat Mass kg	12.6±5.8	12.6±5.5	11.6±3.9	11.4±4.2
FFM kg	58.2±11.3	58.6±11.3	61.1±10.9	62.3±11.0
% Body Fat	17.5±6.7	17.5±6.7	16.2±5.8	15.9±6.5

Data are mean±SD. n=14 Casein-MOR, n=12 Casein-EVE. There were no significant differences within or between groups.

Table 2. Dietary intake.

	Casein-MOR		Casein-EVE	
	Pre	**Post**	**Pre**	**Post**
Kcal/d	1859±683	1975±658	2076±571	2158±552
CHO g/d	193±77	177±66	204±67	186±61
PRO g/d	119±47	167±49*	142±54	173±46*
Fat g/d	68±28	67±30	77±29	80±27
Kcal/kg/d	27±9	28±8	29±8	30±8
CHO g/kg/d	2.8±1.1	2.6±1.0	2.9±1.0	2.6±0.9
PRO g/kg/d	1.7±0.6	2.4±0.6*	1.9±0.6	2.4±0.5*
Fat g/kg/d	1.0±0.3	0.9±0.3	1.1±0.4	1.1±0.4
Cholesterol mg	304±144	381±222	489±363	505±310
Sodium mg	2484±867	2255±903	3019±938	2664±781
Sugar g	79±42	69±35	54±28	52±31
Fiber g	22±8	20±6	22±6	22±7

Data are mean±SD. *Significant difference (pre vs post, $p < 0.05$). There were no between group differences. Legend: CHO – carbohydrate, g – grams, mg – milligrams, PRO – protein. Note: the values for protein intake includes the protein supplement.

DISCUSSION

The present study examined the effects of nighttime casein supplementation in resistance-trained subjects. In brief, there was no difference in any parameter measured (i.e., body composition or performance) whether supplemental casein protein was consumed in the morning or nighttime. Our study confirms prior work in our lab, which shows that in the absence of a training regimen change, the mere addition of dietary protein has neutral effects on measures of body composition or performance (2, 3).

Table 3. Exercise performance.

	Casein-MOR		Casein-EVE	
	Pre	**Post**	**Pre**	**Post**
Volume load	80216±12281	78482±14218	70324±34334	70659±33542
1-RM BP kg	93.7±47.5	95.7±45.4	91.6±39.1	90.2±37.0
RTF	27.2±6.2	26.0±4.6	25.1±5.1	26.3±2.8
SUP erg sec	346.4±72.6	299.9±67.6	273.2±50.9	263.3±48.6

Data are mean±SD. There were no pre-post or between group differences. Legend: 1-RM – one repetition maximum, BP – bench press, erg - kg – kilograms, RTF – repetitions to failure at 60% of the Bench Press 1-RM, wk – week, sec – seconds, SUP erg – stand up paddling ergometer. (Volume load n = 24, 1-RM BP n = 16, RTF n =16, SUP erg n = 8). Note: volume load is measured in total weight lifted per week.

Very few studies have examined the effect of nighttime or evening casein supplementation (8, 15). Burk et al. examined what they termed a "time-divided" (TDR) versus a "time-focused" (TFR) pattern of casein ingestion (8). The TDR pattern of supplementation included a single dose of casein protein in the morning followed by a second dose ingested in the evening 5 hours post-exercise. Conversely, the TFR pattern of supplementation included casein protein in the morning and then also pre-workout in the afternoon. The daily dose of the supplement included 70 g of protein (82% casein) and less than 1 g of carbohydrate and fat. Thus, the actual comparison was between pre-workout and 5-hours post-workout (evening) inasmuch as both treatments included morning supplementation of casein. Interestingly, in this investigation they discovered that the TDR but not the TFR pattern of casein ingestion resulted in a significant increase in fat free mass. Inasmuch as both groups (TFR and TDR) consumed 2.2-2.3 g/kg/d of protein, one would posit that it was the timing of casein protein ingestion (i.e., evening ingestion in the TDR group) that elicited the more favorable anabolic response. With regards to muscular strength, both groups improved similarly. It should be noted that this investigation used untrained subjects in contrast to our current study.

Res et al. examined the effects of acute casein supplementation in 16 young males that performed a single bout of resistance-type exercise in the evening (2000 h) after a full day of dietary standardization (15). Thus, this study did not actually measure body composition. Thirty minutes before sleep (2330 h), subjects ingested a beverage with (40 grams of casein protein) or a non-protein placebo. These investigators discovered that protein ingestion prior to sleep increased whole-body protein synthesis rates and improved net protein balance. Moreover, mixed muscle protein synthesis rates was higher under the protein condition (15). Thus, at least in the very short-term, casein ingestion does indeed enhance protein synthesis.

Ormsbee at al. examined the effects of nighttime feeding of carbohydrate or protein combined with exercise training in sedentary obese women (14). For a treatment period of 4 weeks, subjects consumed whey, casein or carbohydrate (140-150 kcal serving) every night of the week within 30 minutes of sleep. They found that exercise training alone increased strength and lean body mass while decreasing body fat. There was no difference between the three groups vis a vis body composition or performance. This is unsurprising in that sedentary individuals will have a robust response to an exercise stimulus; therefore, any additional dietary intervention will be negligible. Furthermore, in contrast to our study using trained subjects, the Ormsbee investigation examined the effects of supplementation in sedentary obese women. Similarly, Snijders et al. discovered that consuming a protein-containing supplement (27.5 g protein, 15 g carbohydrate, 0.1 g fat) prior to sleep increased skeletal muscle cross-sectional area as well as muscular strength(16). However, the subjects in this investigation were not resistance-trained. Furthermore, they did not equate total protein intake between groups.

There are several limitations with our investigation. Our study was in all likelihood under-powered due in part to the preliminary nature of the investigation. Secondly, we did not control the training program of these individuals mainly due to the fact that we wanted to isolate the effects of timed protein supplementation only. Because of the scarcity of evidence regarding nighttime supplementation of casein protein, it is difficult to arrive at any conclusive recommendation. Most prior studies that involved casein supplementation did not address the fact that the mere addition of protein, particularly in untrained subjects, might impact measures of body composition or performance. Meaning that in order to truly test the effects of nighttime casein feeding, one must also include a group that consumes casein during a different time of the day. Until investigators equate total protein intake, then it would be mere speculation as to whether it is total protein intake or protein timing per se that would induce the biological effect. In all likelihood, both play a role. Moreover, the lack of data in trained subjects makes comparisons difficult because other studies have used untrained individuals.

On the other hand, our data suggest that the mere addition of protein calories does not adversely affect body composition. That is, it is difficult to increase fat mass simply by consuming more protein. This is in agreement with previous work (2, 3, 6). Conversely, without a change in one's training regimen, it is highly unlikely that a dietary shift towards a greater protein intake will change performance or body composition(1). Subjects in the current study were consuming sufficient protein at baseline to meet the needs of their current training (9) although their intakes would not be considered "high." On the pragmatic side, there is no harm and a possible benefit from nighttime supplementation of casein protein (8, 15). Future work should examine the effects of casein supplementation (i.e., nighttime versus morning) in subjects who partake in a more rigorous and different exercise-training program. We would speculate that in order for a dietary intervention to have any meaningful impact, it must also be combined with a change in one's training. A dietary intervention of enhanced protein intake in trained subjects in the absence of training alterations will likely have a neutral effect. Though we would speculate that the best practical advice to give athletes is to consume protein pre-sleep.

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