Indian Journal of Fundamental and Applied Life SciencesISSN: 2231– 6345 (Online)
An Open Access, Online International Journal Available at www.cibtech.org/jls.htm
2014 Vol. 4 (3), pp. 677-682/Alipoor et al.
Research Article

THE EFFECT OF LOW FAT MILK AND CALCIUM SUPPLEMENT + VITAMIN D AFTER COMBINED TRAINING ON FAT MASS, STRENGTH AND AEROBIC POWER OF TRAINED WOMEN

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ABSTRACT
This study investigated the combined training effects and milk and calcium supplement + vitamin D consumption on fat mass, strength and aerobic power (vo_{2max}). Trained women $(N = 20)$ participated in a 8-wk combined – training program. The milk group consumed 2 serving of milk and calcium supplement group consumed an isocaloric product with calcium + vitamin D. Strength, body composition, calcium, Serum 25- hydroxyl vitamin D and (vo_{2max}) were measured before and after training. BF% decreased in both groups with a greater decreased in milk group. Strength in the lower body and (vo_{2max}) increased in both groups. Vitamin D and Ca increased in both groups and this increase in vitamin D was significant in milk group $(P = 0.027)$. It can be acknowledge that milk is a suitable drink for favorable change in body composition with combined training in women.

Keywords: *Body Fat, Fat Mass, Strength, Aerobic Power, Combined Training*

INTRODUCTION
Combined exercise is a potent stimulus to increase muscle protein synthesis (Josse *et al.,* 2010). Resistance training maximizes hypertrophic potential and it appears that consumption of high- quality protein can augment muscle protein synthesis (Fujita *et al.,* 2007).

Previous studies have reported that women engaging in resistance training for as little as 6 wk show comparable relative gains in strength as those seen in men (Staron *et al.,* 1991).

Resistance training is not a common exercise modality of choice in young women, however, the health benefits they stand to gain from this form of exercise in terms of improving physical strength and muscular cannot be achieved by other means (Chilibeck *et al.,* 2002; Williams *et al.,* 2007). Reports indicate that milk is a suitable material in loss of fat mass increasing muscular mass.

Consuming dairy products such as milk after a resistance training program provide protein and carbohydrate, necessary macronutrients to maximize the anabolic response to resistance exercise (Tipton *et al.,* 2004). Milk contains both calcium and vitamin D.

These micronutrients might further enhance the benefits of combined exercise because of their associations with fat oxidation (Teegarden *et al.,* 2008). Therefore, the objective of this study was to determine the effect of low- fat milk and calcium supplement consumption after to 8-week combined training (Aerobic- resistance) program on loss fat mass, strength and maximal oxygen consumption (vo_{2max}).

MATERIALS AND METHODS

Methods

Trained women (n = 20, age 19 – 30) were recruited. Inclusion criteria included baseline habitual low-calcium ($\leq 800\ mg\ /day\ total$) and low- dairy ($\leq 1 serving\ /\ day$) diet, participation in aerobic and resistance training ≥ 5 hr/week. Exclusion criteria included high blood pressure, lactose intolerance, lactation or pregnancy, digestive or metabolic diseases, diabetes, use of medication that might interfere with calcium absorption. Calcium intake was initially assessed and participants completed the physical activity readiness questionnaire (PAR-Q).

Twenty subjective were randomly assigned to two groups: milk group (MILK) and Ca+ vit D group, with n = 10 in each. Subjects consumed 500 ml of low- fat milk (1.5% fat) (250 mg Ca+ 100 IU vit D) on two occasions (immediately and 1 h after exercise) daily (totaling 1 L of milk perday) or calcium supplementation + vit D (500 mg Ca+ 200 *IU* vit D).

The study protocol consisted of 8 wk, 3d/ wk, of combined exercise training included 10 min warm up, 5 min stretching, 30 min aerobic and 40 min resistance exercise and 5 min cool down. Briefly, the training regimen consisted of: (45° incline leg press, seated two- leg hamstring curl, seated two- leg knee extension, seated calf raise, seated lateral pull down, chest fly, triceps, biceps push down, and abdominal exercise without weight) all exercise were performed at 60% of the subject's voluntary single repetition maximal strength (1 RM) with three sets of 15 repetitions of each exercise were performed with a 2- min rest between sets. Weight was recorded in light clothing to the nearest 0.1 kg, using a digital, scale; height was measured bare foot to the nearest 0.1 cm using a stadiometer. Then BMI calculated as weight (kg)/ height waist and hip circumferences were measured to the nearest 0.1 cm using a plastic measuring tape at the smallest circumference below the rib cage and above the umbilicus (Josse *et al.,* 2010) and body fat was measured at the beginning of the study and at week 8 using saehan caliper model by using the equation of Pollock & Jackson (Shapes *et al.,* 2004)

Assessment

Strength

A successful 1 RM was when the subject was able to move the machine through a full range of motion. The subject was allowed a full 3- min rest before attempting a second lift.

Maximal Oxygen Consumption (vo_{2max})

One of the aerobic fitness test is "maximum oxygen uptake (vo_{2max})" that in order to aerobic power, athlete's vascular preparedness is measured. One of these tests is the Bruce test. Bruce running test estimates maximum oxygen consumption with time.

Biochemical Measurements

25 – Hydroxyl vitamin D (25 OHD) concentration was measured in serum obtained from blood samples at baseline and after the 8- week training program via a 25 OHD radioimmunoassay (RIA). 25- hydroxyl vitamin D levels less than 16.6 mg/ml in women was considered as the shortage criterion. To measure the amount of serum ionized calcium, ionized calcium electrode device of ion selective type with KONE brand limited states of America construction, were used.

Data were analyzed using spss (version 18.0.1). Descriptive data from baseline by group were analyzed using one- way analysis of variance (ANOVA). Two- way, repeated- measures ANOVA with time (before and after) as the within –subjects factor and group (milk and Ca + vit D) as the between –subjects factor were carried out for all body composition variables, dietary variables, strength, (vo_{2max}) changes, prestudy and post- study measures.

RESULTS AND DISCUSSON

Results

Table 1: Baseline characteristics of subjects under study

Variable	Milk group (n=10)	Ca group (n=10)	P – value
Age (year)	24.67 ± 2.57	23.50 ± 4.72	0.52
BMI (kg/m^2)	25.79 ± 2.57	25.79 ± 1.86	0.10
WHR	0.87 ± 0.04	0.87 ± 0.04	0.41
Body fat (%)	31.13 ± 4.33	30.58 ± 2.49	0.75
Calcium (mg/d)	9.11 ± 0.26	9.11 ± 0.48	0.18
25- hydroxyl vitamin D (mg/ml)	9.22 ± 2.41	14.10 ± 4.88	0.18
(vo_{2max})	44.48 ± 7.36	43.51 ± 6.02	0.77
Strength (1RM (lower body)	72.53 ± 32.45	75.100 ± 39.13	0.58
Strength (1RM (upper body)	31.91 ± 13.69	29.32 ± 11.87	0.51

** P < 0.05 is significant*

Table 2: Changes of body composition pre-post training

	Milk			Ca		
Variable	**Pre**	**Post**	**p. value**	**Pre**	**Post**	**p. value**
BM (kg/m^2)	25.79 ± 1.86	22.19 ± 1.22	0.001	25.79 ± 1.86	24.08 ± 1.77	0.003
WHR	0.87 ± 0.04	0.85 ± 0.03	*NS*	0.87 ± 0.04	0.87 ± 0.05	0.06
Body fat (%)	31.13 ± 4.33	22.18 ± 3.40	0.00	31.13 ± 4.33	24.8 ± 6.05	0.002

[1] *All values are* $\bar{X} \pm$ *SD statistical was by 2- factor ANOVA (pre and post) and between (milk or Ca + vit D) variables with (ANCOVA) test.*
[2] *Significantly different from MILK and Ca + vit D at the same time point P < 0.05*
[3] *Significantly different from pre and post within and post within the same group P<0.05*
[4] *NS: non- significant*

Table 3: Intra – Inter group changes in the subjects

	Milk	Ca				
Variable	**Pre**	**Post**	**p.value**	**Pre**	**Post**	**p. value**
Calcium	9.11 ± 0.48	9.53 ± 10.39	*NS*	9.11 ± 0.48	9.18 ± 0.22	*NS*
25-hydroxyl vitaminD	9.22 ± 2.41	14.76 ± 5.34	0.027	14.10 ± 4.88	14.18 ± 10.39	*NS*
VO_{2max}	44.48 ± 7.36	49.22 ± 8.09	0.00	43.51 ± 6.03	48.69 ± 6.56	0.061
Lower body strength (1RM)	72.53 ± 32.45	93.05 ± 47.34	0.055	75.100 ± 39.13	90.09 ± 44.31	0.02
Upper body strength (1RM)	31.91 ± 13.69	36.08 ± 16.53	0.98	29.32 ± 11.87	48.34 ± 2.06	*NS*

* $p < 0.05$ *is significant.*

Research Article

Comparing the characteristics of participants, including age, body fat, WHR, Ca and 25- hydroxyl vitamin D, (vo_{2max}) and strength (1RM), at the beginning of research suggested that there were no significant difference between subjects in two groups.

According to table2, the amount of body fat (%), WHR, BMI after 8/w of intervention in both of groups, showed a significant decrease (P<0.05), but the difference in these changes between two groups only for the variables of BMI and % BF were significant.

According to table 3- calcium level in both milk and calcium groups increased and so amount of 25- hydroxyl vitamin D in milk group after the intervention increased significantly (P= 0.027).

Lower body strength in milk group increased significantly (P=0.055). This increase was significant in the calcium group as well (P=0.02).

Upper body strength in milk group had a significant increase (P=0.98), but this increase was not significant in the calcium supplement group.

Discussion

The result of this study demonstrated a significant reduction in weight, BMI, WHR, body fat (%) in the calcium supplement and milk groups, our findings also indicate that a milk consuming can lead to a higher reduction in BMI and BF%. Zemel *et al.,* (2004) showed that 18 subjects receiving 3 cups of yougurt per day (1100 mg/day calcium) lost more weight (6.63 kg vs. 4.99 kg), body fat (4.43 kg vs. 2.75 kg), while attenuating loss of fat-free mass (1353 g vs. 19 689) in comparison with 16 subjects in a low Calcium control group (400 – 500 mg/day). Zemel *et al.,* (2005) in two randomized studies African-American women with obesity compared the subjects under weight loss diet with those who were under the weigh- balanced diets, in the conditions of receive high or low dairies, and observed that calcium- rich food leads to greater weight loss in these individuals. In another study Fagih *et al.,* (2011) compared the effects of low-fat milk consumption, soy milk fortified with calcium and calcium supplement on weight and body fat loss in over weight and obese premenopausal women, and the comparison of weight changes average. Waist circumference, BMI and WHR showed that the reduction in waist circumference and WHR among four group control, calcium supplement, soy milk and milk had a significant difference and of course, these change in group consuming milk were more than other groups that is clinically important Zemel *et al.,* (2004) also studied the effect of an energy- restricted diet (providing either 400- 500 mg/day from dairy products or 1200-1300 mg/day from an additional 800 mg of calcium carbonate or from an additional three servings of dairy products) on weight and fat loss in 32 obese or overweight women. Their results indicated that increasing dietary calcium significant augmented weight and fat loss secondary to caloric restriction.

Hartman *et al.,* (2007) as well in a study to determine the effect of fat- free milk consumption after resistance training compared to the isoenergetic soy or carbohydrate- only beverages in young men athletes, observed greater decreases in FM in untrained men (n=56) who consumed fat-free milk post exercise than in those who consumed isoenergetic soy or carbohydrate- only beverages. Also Shapese *et al.,* (2004) during the 25 weeks of the effect of 100 mg calcium on 100 premenopausal and postmenopausal women, observed that there was no significant change in weight or fat mass between calcium supplement and placebo groups. Also Barr *et al.,* (2003) in a review of 17 studies on the effects of dairy consumption and calcium supplementation on osteoporosis, concluded that receiving dairy had no significant effect on body weight or fat mass following the consumption of milk and calcium supplementation after resistance training. The findings of our study indicate that the calcium and vitamin D amount has increased significantly in both groups and this increase of vitamin D in the milk group (P <0.027) was meaningful and these findings are consistent with Zemel *et al.,* (2000) examination which the effects of calcium enhancing with oral intake of dairy origin calcium was more remarkable by adding two cups of low- fat yogurt to diet and compare it with effects of calcium supplement. On the other hand, calcium increase is correlated with decreasing of body fat percentage. On the other hand Morris and

Zemel (2005) showed that 1.25 dihydroxy vitamin D is directly regulated 11- hydroxysteroid hydrogenase 1 expression in adipocytes.

Research evidence show the effect of calcium supplementation and milk on strength, in lower body part in milk and calcium supplement group increased significantly which this was not compatible with the study results of Josse *et al.*, (2010), so that in the study. The muscle strength in the bench press and triceps in milk group was more than that of carbohydrate with the same calories. Hence, consumption of milk after resistance training against the carbohydrates with the same calories, will lead to further growth of muscle mass, fat mass loss and strength gain after 12 weeks of resistance training in women. But in the present study, according to the sporting history of study participants, and given that not exercised women have half the strength in the upper body and about 2-3 times more in lower body compared to not exercised male competitor, so the possible cause of strength surge in women consuming milk, reflects more potential in women due to the more primary strength in the lower body.

Also in the study of White *et al.*, (2009) with research on changes in body composition by eating yogurt during resistance training in women with chest and crus strength test, concluded that subjects in yogurt group had more power than the carbohydrate group or proteins group. About the effect of milk and calcium supplementation on vo_{2max}, after the intervention it can be stated that there is no significant difference in milk (p=0.000) and calcium supplement (p=0.001) groups.

Since the oxygen consumption is expressed absolutely in terms of its relation to net weight (fat free) of body, when vo_{2max} is expressed in connection with body weight (ml/kg/minute), the difference will more based on fat free body weight. In the present study, considering that the BMI reduction in milk group was significantly higher than the calcium group, and fat mass percentage in milk group was lower than the calcium group, naturally (vo_{2max}) amount had a significant increase compared to calcium group. Thus it can be concluded that participation in combined training have been effective in increasing the performance time on treadmill in subject Bruce test.

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