



## Effect of milk protein concentrate supplementation on body composition and biochemical markers during a resistance training program

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### Abstract

This study aimed to formulate a milk protein concentrate (MPC) and evaluate the effect of this MPC supplementation on body composition, lipid profile, and product acceptability in resistance training (RT) practitioners. The physical, chemical and sensory properties were then analyzed. To evaluate the effect of the MPC supplementation on resistance training practitioners, 30 males were divided into two groups: a control group and an intervention group. Then the participants in the intervention group were supplemented with 500 mL of the MPC (250 mL pre-RT and 250 mL post-RT). Body composition (weight, height, and skinfolds) and biochemical markers (cholesterol and triglycerides) were measured at baseline and at 30, 60, and 90 days after treatment. Body composition results showed that the MPC supplementation during a RT program significantly ( $p < 0.05$ ) reduced body fat percentage and fat mass, and increased lean mass in RT practitioners. Characterization of the MPC revealed low fat ( $3.32 \pm 0.08\%$ ) and carbohydrates ( $4.58 \pm 0.59\%$ ), and high protein ( $18.51 \pm 0.51\%$ ) concentrations. Additionally, the product showed satisfactory acceptability index ( $> 71\%$ ). Thus, the MPC supplementation contributed to reduction of fat percentage, fat mass, and increase of lean mass in RT practitioners, without lipid profile alterations.

**Keywords:** strength training; dairy products; supplements; nutrition; anthropometry.

**Practical Application:** This work shows that it is possible to develop a health safe protein drink with good acceptability for RT practitioners which can positively influences the body composition parameters.

## 1 Introduction

Resistance training (RT) is becoming increasingly popular; due to benefits such as neuromuscular and body composition improvements (American College of Sports Medicine, 2009). In addition, RT promotes the reduction of risk factors for diseases such as coronary heart disease and diabetes, improvement of cardiorespiratory capacity, prevention of osteoporosis, reduction in the risk of colon cancer, and improvement of mental health (American College of Sports Medicine, 2009). Most people who practice RT consume food supplements, with the aim of promoting muscle development and performance (Dias et al., 2005). Supplements help complement the diet of a person and are ergogenic resources used to improve performance in sports and fitness activities (Moreira et al., 2013)

The popularity of supplements is directly linked to people seeking healthy and nutritious food options that help improve physical fitness (Cavalheiro et al., 2020). The supplements most consumed by RT practitioners are protein supplements, particularly albumin and whey proteins (WP) (Velasco et al.,

2022), obtained after the removal of casein from milk, and contain a large amount of amino acids, calcium, and bioactive whey peptides (Alves & Lima, 2009). Proteins from milk whey are rapidly digested and absorbed rapidly increasing plasma amino acid levels after ingestion; whereas casein is slowly absorbed, and decreases the stimulus of protein synthesis after ingestion (Souza et al., 2015).

In a recent study, Huschtscha et al. (2021) reported positive effects of milk protein concentrate (MPC) in older adults but no effects on systemic inflammatory cytokines, hormonal responses, body composition, and physical performance after a 12-week RT program. Antonio et al. (2016) observed that a 4-month RT period had no effect on blood lipid levels, renal markers, hepatic parameters, physical performance, or body composition in young men. However, Pourabbas et al. (21) showed that the use of MPC during six weeks of RT significantly improved lean mass, strength, power, and altered serum concentrations of skeletal muscle regulatory markers in trained young men.

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One of the great challenges for food development is to guarantee a good acceptability of the product. In general, the acceptability of a food and its commercial success depend directly on its sensory attributes (Siqueira et al., 2013). Therefore, it is essential to carry out studies related to the sensory issue and the acceptability of the product by consumers during its development (Pinto et al., 2022; Pinto et al., 2019). In addition, an evaluation of the functionality and benefits related to its consumption, such as mass gain, are of great value when seeking to develop products for practitioners of physical activities. Several products have been developed in this context, such as protein cereal bars (Jabeen et al., 2021a; Jabeen et al., 2021b), protein yogurts (McKinlay et al., 2022), protein foods of plant origin for vegans (Naclerio et al., 2021), among others.

In this context, there are gaps in the literature about the effects of MPC supplementation in RT practitioners with a product that combines yogurt and ricotta, replacing WP, commonly found in the market, to assess possible benefits to health and body composition. Therefore, the present study aimed to develop an MPC and evaluate the effect of its supplementation of body composition, lipid profile, and product acceptability in RT practitioners.

## 2 Materials and methods

### 2.1 Participants

The study was carried out with thirty male participants, 15 in control group (CG) with a mean age of  $22.2 \pm 4.9$  years and 15 in intervention group (IG) with a mean age of  $28.7 \pm 8.8$  years supplemented with 500 mL MPC (250 mL pre-RT and 250 mL post-RT). The inclusion criteria were males  $\geq 18$  years of age, who practiced RT for at least 6 months, did not have food allergies or intolerance, and did not have diabetes, gastrointestinal, or cardiovascular disease. The exclusion criteria were pathologies or chronic diseases such as obesity, hypertension, and diabetes, consumption of food supplements in the 60 days before the start of the study, consumption of alcohol, illicit drugs, antibiotics, and smoking. The anamnesis form was used for participant evaluation.

### 2.2 Ethical details

The study was conducted in accordance with the Declaration of Helsinki guidelines. All participants read and signed a consent form, and the study was approved by the local ethics committee (protocol number 3.248.372).

### 2.3 Procedures

The participants in the IG group were supplemented with the MPC during 90 days of RT (six sessions per week). RT was performed in both groups, with four exercises for each muscle group, (four sets of eight repetitions (American College of Sports Medicine, 2009) and 2 minutes interval between sets and exercises (Silva et al., 2019) and intensity of 80% maximal repetition (1RM) (Simão et al., 2007). 1RM tests and re-tests were performed according to the protocol proposed by Simão et al. (2022). To assess the effects on body composition and biochemical markers during training in both groups, anthropometric

assessment (weight, height, and skinfolds) and blood analysis (total cholesterol, HDL cholesterol, and triglycerides) were performed at baseline and 30, 60, and 90 days after training.

### 2.4 Formulation of MPC

In natura milk was obtained from the milking of crossbred cows at the Laboratory of Bovinocultura Leiteira, Goiano Federal Institute, Campus Rio Verde, GO, Brazil. The milk was transported in polyethylene drums (20.0 kg capacity) to the Laboratory of Animal Origin Products (LPOA) of the Goiano Federal Institute, Campus Rio Verde, GO, Brazil. For the production of whole yogurt, the milk was filtered, heated to a temperature of 60 °C and 10% sucrose was added to it, followed by pasteurization at 90 °C for three minutes. This mixture was then cooled to 42 °C, inoculated with 10% lactic yeast (*Lactobacillus bulgaricus* and *Streptococcus thermophilus*), and homogenization for three minutes. Subsequently, the mixture was placed in sanitized and capped glass flasks, incubated at 42 °C, kept at rest until reaching pH 4.6. The pH was controlled by addition of aliquots regulated using a pH meter (NOX68, Noxtron). Subsequently, it was cooled to 20 °C, resulting in gel breakage and cessation of fermentation. The resulting yogurt was then stored at a temperature of 5.0 °C, as recommended by Krolow & Ribeiro (2006).

To produce ricotta from skimmed milk, fresh milk was filtered, heated to a temperature of 40 °C, and then skimmed to obtain skim milk and cream. The skimmed milk was pasteurized at 72 °C for 20 s, then the temperature was reduced to 60 °C, with the addition of 0.3% citric acid to the total volume of milk, with light agitation until the mass flocculated. Then the ricotta mass was removed, filtered for 60 s, after which the ricotta was stored at 5 °C. The ideal formulation of MPC for supplementation was carried out in the laboratory. Homogenization and texture was visually evaluated and final protein concentration of the milk concentrates was estimated. The following formulations were evaluated: 60.0% of ricotta + 40.0% of yogurt, 70.0% of ricotta + 30.0% of yogurt, 80.0% of ricotta + 20.0% of yogurt, and 90.0% of ricotta + 10.0% yogurt. The MPC was then packed in glass containers and refrigerated at 7 °C to be distributed to the analysis assessors. For the production of the MPC used in this study, 500 liters of in natura milk was required for the production of whole yogurt and 5000 liters of skimmed milk was required for the production of ricotta. At the end of the study, 675 kg of MPC was produced.

### 2.5 MPC chemical and physical analysis

The proportion of fat, protein, lactose, defatted dry extract (DDE), and total dry extract (TDE) of the in natura milk was analyzed using Milkoscan 4000 (Foss Electric A/S. Hillerod, Denmark) based on the differential absorption of infrared waves by different components of milk. The samples were heated in a water bath at 40.0 °C for 15 min for fat dissolution. The results were expressed as percentages (%) (International Organization for Standardization, 2013). The casein content was determined using Fourier-transform infrared spectroscopy (Lactoscope equipment, Delta instruments, United States). The concentration of casein were expressed as percentages (%). Somatic cell count (SCC) analysis was performed using Fossomatic 5000 flow cytometer (Foss

Electric A/S. Hillerod, Denmark). Before analysis, the samples were heated in a water bath at 40.0 °C for 15 min to dissolve fat. The results are expressed as SCC per mL (International Organization for Standardization, 2006). The total bacterial count (TBC) was analyzed using Bactoscan FC flow cytometer (Foss Electric A/S. Hillerod, Denmark). The results were expressed as UFC per mL (International Organization for Standardization, 2020).

To evaluate the fat in the MPC, the Gerber method [Normative Instruction no. 68, December 12, 2006 (Brasil, 2006)]; total nitrogen by the micro-Kjeldahl method [official standard No. 960.52, the Association of Official Analytical Chemists (2005)], multiplied by the conversion factor 6.38 for dairy products, to obtain the protein content; ashes by means of total carbonization of organic matter in a muffle furnace (Quimis®) at 550 °C for 8 h, until light ash was obtained [official method No. 923.03, the Association of Official Analytical Chemists (2005)]; water content using gravimetry by heating in an oven (Thoth 250 L) at 105 °C for 24 h, [official method No. 925.10, the Association of Official Analytical Chemists (2005)]. Concentration of carbohydrates was estimated by the difference between the total sample (100.0%) and the moisture, protein, lipid, and ash contents, expressed in percentage. The calorific value was calculated using the Atwater coefficient (Torres et al., 2000). The pH of the samples was measured directly using a pH meter (NOX, Noxtron).

## 2.6 Sensory analysis

To assess the acceptance of the MPC, sensory analysis was performed individually in the morning (9:00 to 10:00) and afternoon (17:30 to 19:00). The evaluators received a form and a sample in a 50 mL white disposable plastic cup containing 30 g of sample at 7.0 °C. The tasters were asked to indicate the acceptance of the product on a structured hedonic scale of nine points, with 9 being “extremely liked” and 1 “extremely disliked”. The acceptability index (AI) was calculated according to Monteiro (1984), using the following Equation 1:

$$AI(\%) = \left( \frac{A}{B} \right) 100 \quad (1)$$

AI, acceptability index; A, average score obtained for the product; B, maximum grade given to the product.

Then the evaluators received a form in which they were asked to indicate the intention to purchase the product on a hedonic scale with a score from 1 to 5 with a score of 1 indicating “I would certainly not buy the product” and a score of 5 indicating “I would certainly buy the product” (Minim, 2013).

## 2.7 Anthropometric assessment

The anthropometric parameters of body weight and height were assessed following the protocol of Fernandes (2003). A clinical scale (Filizola) and the body mass index (BMI) was calculated using the Equation 2.

$$BMI = \frac{Weight(kg)}{Height(cm)^2} \quad (2)$$

The percentage of subcutaneous fat was determined from the following skinfolds: suprailiac, subscapular, triceps, abdominal, middle axillary, thigh, and pectoral, and measurements were performed by a single evaluator, to avoid possible errors (Jackson et al., 1980). Physical parameters were assessed at baseline and at 30, 60, and 90 days after RT.

## 2.8 Biochemical analysis

Biochemical tests for lipid assessment were performed at the Hormonal Laboratory of the City of Santo Antônio da Barra (GO). Triglycerides and total cholesterol were measured using Spectrum CCX II calibrated with TruCal (Abbott Diagnostics, Abbott Park, IL, USA) as per manufacturer’s specifications (standards: using Triglycerides FS (DiaSys Diagnostic Systems GmbH & Co. KG, Holzheim, Germany), and cholesterol (BioSystems S.A., Barcelona, Spain)). The tests were calibrated with the CCX Multicalibrator Set (Abbott) with three-point curves. HDL-C dosage was determined using a homogeneous method without precipitation using the HDL-C Immuno FS reagent as per manufacturer’s instruction.

Blood samples for lipid profile assessment were collected 72 hours before starting the RT and MPC supplementation, and after ending the RT and MPC supplementation at 72 hours.

## 2.9 Statistical analysis

For the statistical evaluation of the data from the physicochemical and sensory analyses (test of acceptability and purchase intention), Sisvar 5.6 software was used. The means were compared using the Tukey test. The RT practitioners were distributed in a completely randomized design and divided into two groups, control and intervention, during 90 days of training and supplementation with MPC.

Initially, descriptive statistics, mean values, and standard deviations were used to record the characteristics of the participants. The Kolmogorov-Smirnov test was used to verify the normality distribution of the data. The reliability of the 1RM test and re-test sessions was analyzed using a two-factor random effect model intraclass correlation coefficient (ICC) and paired *t*-tests. To compare the groups (control vs. intervention) and to study the changes in parameter with time (baseline and at 30, 60, and 90 days after RT), analysis of variance (ANOVA) test of repeated measures with post hoc analysis (Sidak) was used for variables that showed normal distribution. The intergroup comparison at each time point was verified using the Friedman test, and the comparison between groups was done using the Kruskal-Wallis test for the variables that without a normal distribution.

The differences between the groups post- and pre-intervention (delta) was evaluated using the Student’s *t*-test or its nonparametric equivalent, the Mann Whitney U test. Anthropometric data were analyzed using the Statistical Package for Social Sciences (SPSS, version 20.0; SPSS Inc., Chicago, IL, USA). A significance level of 5.0% was used for all analyses.

### 3 Results

The 1RM values showed excellent reproducibility between the test and re-test ( $ICC > 0.90$ ), and the paired  $t$ -test did not demonstrate significant differences ( $p > 0.05$ ). The formulation with 70.0% ricotta and 30.0% yogurt, homogenized in a domestic blender for 4 min, until complete homogenization was identified as the best formulation for processing MPC. Tables 1-3 showed physicochemical properties of whole and skimmed milk, full-fat natural yogurt, skimmed ricotta, and MPC; the MPC for RT practitioners acceptability indices. Tables 4-5 report the effect of supplementation on the physiological profile of the participants and on their body composition between the post-test and pre-test, respectively.

Mean fat values differed significantly ( $p < 0.05$ ) between skimmed and whole milk. Despite the average variation, the protein results did not differ significantly ( $p > 0.05$ ) between the two types of milk (whole and skimmed). Lactose, TDE, DDE, and SCC values differed significantly ( $p < 0.05$ ) between milk types. No significant differences ( $p > 0.05$ ) were observed in the TBC analysis, despite the significant differences in the types between milk types. The mean values of fat, protein, carbohydrate, ash, DM, water content, and pH differed significantly ( $p < 0.05$ ) between products (yogurt vs. ricotta vs. MPC). The calorific value of MPC differed significantly ( $p < 0.05$ ) only in yogurt.

Comparing the groups of RT practitioners (control vs. intervention), no significant differences ( $p > 0.05$ ) were observed in the appearance, color, aroma and texture of the MPC, despite the significant differences ( $p < 0.05$ ) of taste, global assessment and purchase intent. The participants of the control group

suggested the following changes in the product: addition of fruit (6.67%), addition of sugar (13.33%), reduction in viscosity (20.0%), changing the taste (13.33%), in addition of flavoring and sweetening agents (33.33%), reduction of grittiness (13.33%). If the product was flavored, the preferred flavors would be banana (6.66%), chocolate (40.0%), coconut (20.0%), red fruits (6.67%), and strawberry (26.66%).

In the intervention group, 46.67% reported that they would change the flavor, followed by a decrease in grittiness (20.0%), addition of sugar (20.0%) and a decrease in consistency (6.67%), and decrease in acidity (6, 67%). The preferred flavors were strawberry (46.67%), chocolate (33.33%), and vanilla (20.0%).

### 4 Discussion

#### 4.1 Biochemical and anthropometric evaluation of MPC consumption

Our results showed that the mean values of fat percentage differed in both groups, being significantly lower in the IG group (Table 1). In addition, the fat percentage during this study in the IG showed a reduction after 30 days of intervention, while in the CG, the reduction occurred only after 60 days, demonstrating that the supplementation of MPC during a RT program can bring improvements for consumers, reducing the percentage of fat. Supplementation was also effective in increasing lean mass and reducing fat mass.

The percentage reduction in fat content in our study was higher than that reported in the literature. Antonio et al. (2016) evaluated the effects of a protein-rich diet on the body composition of RT practitioners and reported no significant differences between the

**Table 1.** Mean values and standard deviation of body weight, body mass index (BMI), fat percentage, lean mass, fat mass and lipid profile (total cholesterol, triglycerides and high density lipoprotein - HDL) of RT practitioners.

Variables	CG (n = 15)				p-value
	Baseline Mean $\pm$ SD	30 days after Mean $\pm$ SD	60 days after Mean $\pm$ SD	90 days after Mean $\pm$ SD	
Body weight (kg)	75.0 $\pm$ 16.9 <sup>a</sup>	74.9 $\pm$ 16.1 <sup>a</sup>	75.1 $\pm$ 15.8 <sup>a</sup>	75.2 $\pm$ 15.3 <sup>a</sup>	0.90
BMI (kg/m <sup>2</sup> )	24.2 $\pm$ 4.3 <sup>a</sup>	24.2 $\pm$ 4.1 <sup>a</sup>	24.3 $\pm$ 3.9 <sup>a</sup>	24.3 $\pm$ 3.8 <sup>a</sup>	1.00
Fat %	18.9 $\pm$ 7.7 <sup>a</sup>	18.6 $\pm$ 7.7 <sup>a</sup>	17.3 $\pm$ 7.7 <sup>b</sup>	16.6 $\pm$ 7.2 <sup>c</sup>	< 0.05
Lean mass (kg)	59.7 $\pm$ 8.6 <sup>c</sup>	60.0 $\pm$ 8.3 <sup>c</sup>	61.2 $\pm$ 8.4 <sup>b</sup>	62.0 $\pm$ 8.5 <sup>a</sup>	< 0.05
Fat mass (kg)	15.3 $\pm$ 9.3 <sup>b</sup>	14.9 $\pm$ 9.0 <sup>b</sup>	14.0 $\pm$ 8.8 <sup>a</sup>	13.3 $\pm$ 8.1 <sup>a</sup>	< 0.05
Total cholesterol (mg/dl)	175.8 $\pm$ 40.5 <sup>a</sup>	NM	NM	167.9 $\pm$ 33.7 <sup>a</sup>	0.10
Triglycerides (mg/dl)	78.1 $\pm$ 19.2 <sup>a</sup>	NM	NM	77.7 $\pm$ 19.3 <sup>a</sup>	0.11
HDL (mg/dl)	45.9 $\pm$ 8.4 <sup>a</sup>	NM	NM	45.4 $\pm$ 7.8 <sup>a</sup>	0.90
Variables	IG (n = 15)				p-value
	Baseline Mean $\pm$ SD	30 days after Mean $\pm$ SD	60 days after Mean $\pm$ SD	90 days after Mean $\pm$ SD	
Body weight (kg)	78.7 $\pm$ 13.4 <sup>a</sup>	79.3 $\pm$ 12.3 <sup>a</sup>	80.1 $\pm$ 11.4 <sup>a</sup>	80.7 $\pm$ 11.1 <sup>a</sup>	> 0.09
BMI (kg/m <sup>2</sup> )	26.5 $\pm$ 4.6 <sup>a</sup>	26.8 $\pm$ 4.3 <sup>a</sup>	27.0 $\pm$ 3.9 <sup>a</sup>	27.1 $\pm$ 3.8 <sup>a</sup>	> 0.10
Fat %	22.9 $\pm$ 5.9 <sup>a</sup>	20.7 $\pm$ 6.3 <sup>b</sup>	18.7 $\pm$ 6.5 <sup>c</sup>	16.8 $\pm$ 6.1 <sup>d</sup>	< 0.05
Lean mass (kg)	60.3 $\pm$ 7.3 <sup>d</sup>	62.4 $\pm$ 7.1 <sup>c</sup>	64.7 $\pm$ 6.8 <sup>b</sup>	66.7 $\pm$ 6.5 <sup>a</sup>	< 0.05
Fat mass (kg)	18.6 $\pm$ 7.6 <sup>a</sup>	17.2 $\pm$ 7.4 <sup>ab</sup>	15.5 $\pm$ 7.4 <sup>bc</sup>	14.0 $\pm$ 7.0 <sup>c</sup>	< 0.05
Total cholesterol (mg/dl)	160.7 $\pm$ 28.7 <sup>a</sup>	NM	NM	156.0 $\pm$ 29.7 <sup>a</sup>	1.03
Triglycerides (mg/dl)	78.1 $\pm$ 30.4 <sup>a</sup>	NM	NM	79.1 $\pm$ 25.3 <sup>a</sup>	1.00
HDL (mg/dl)	46.4 $\pm$ 8.6 <sup>a</sup>	NM	NM	48.6 $\pm$ 8.1 <sup>a</sup>	0.90

SD: standart deviation; NM: not measured. Equal letters (a, b, c, d) indicate similarities in intergroup measures.

evaluated groups (CG vs. IG), demonstrating the effectiveness of specific MPC supplementation adopted in the present study. Perhaps the authors (Antonio et al., 2016) did not find significant differences due to the lower weekly RT frequency compared to our study. Corroborating our study, Pourabbas et al. (2021) observed significant differences in lean mass in RT-trained men. The mean lean mass of the evaluated groups differed from each other (Table 1). Because of the increase in lean muscle mass, the percentage of fat was reduced. We also observed a reduction in body fat in the CG; however, the reduction of fat mass of the IG more as compared to that of the CG. This reduction in both groups may be directly associated with the benefits of RT (American College of Sports Medicine, 2009).

The consumption of MPC by practitioners of RT during 90 days of study did not result in a significant increase in body weight in the evaluated groups (Table 1). Similar results to this research were reported by Antonio et al. (2017) who evaluated casein protein supplementation in participants. The results of this study suggest that the intake of a milk-based protein supplement by RT practitioners is beneficial for changes in body composition. The mean BMI also did not differ between the groups evaluated (Table 1); however, BMI does not consider the amount of lean mass. Our results were similar to those of Panta et al. (2017), who evaluated the effects of personalized RT on the body composition of adult men and reported mean BMI values of 24.30 kg/m<sup>2</sup>. However, Neves et al. (2015) evaluated the effects of RT on the body fat percentage index in untrained adults and found average BMI values of 31.9 kg/m<sup>2</sup> in the first physical evaluation, with an average value of 30.3 kg/m<sup>2</sup>, demonstrating that there is no defined tendency due to several factors influencing the responses, especially the characteristics of the participants, manipulation of RT methodological variables, and duration of the program (American College of Sports Medicine, 2009; Simão et al., 2007; Simão et al., 2022).

In the study by Pourabbas et al. (2021), 6 weeks of linear RT with 4 sessions per week significantly changed the strength, power, and serum concentrations of skeletal muscle regulatory markers in trained young men. The author use a MPC dose similar to our study. Huschtscha et al. (2021) measured body composition, strength (1RM), power, VO<sub>2</sub>, physical performance, and biochemical variables such as immune and hormonal responses (during a 12-week RT program (baseline, 6, and 12 weeks) in elderly men. Despite a significant increase in strength, the authors did not find any differences (CG vs. IG) in performance and biochemical responses. These differences were probably due to the age differences between the samples. Our data also show benefits of MPC during an RT program in men, with no significant differences in biochemical responses (lipid profile).

The lipid profiles showed no significant differences between any of the groups evaluated (Table 1). This was different from the observations of Albarello et al. (2017) which reported a significant difference for triglycerides and HDL before and after RT. Although our data did not show positive differences in cholesterol and triglycerides, the consumption of the milk-based protein supplement by RT practitioners suggested that the product could be consumed to improve in the body composition. Moreover, the study suggests that the MPC did not have any

negative health effect. Thus, in this work, body composition and lipid profiles were compared within each group throughout the supplementation period. Moreover, differences in post- and pre-intervention (delta) measurements were compared between groups, and the body fat percentage, lean mass, and fat mass were significantly greater in the IG than in the CG (Table 2).

The percentage of fat mass in both groups was reduced, however, the reduction was not significant (Table 1). However, the reduction in the percentage of fat in the CG was -2.30% and of that IG -6.08% (Table 2). The lean mass of the studied groups did not show significant changes as shown in Table 1, however, it was observed that the highest increase in the lean mass was reported (Table 5) in the IG with an increase of 6.40 kg; while that in the CG was only 2.12 kg. Similarly, the highest reduction was observed in IG (4.5 kg) as compared to the CG (1.91 kg). Thus, we observed that RT is associated with improvements in the body composition changes (an important component of health-related physical fitness) and that MPC contributes to this improvement.

New studies involving other RT methods, biochemical markers, physical tests, and neurophysiological variables are suggested to understand the real effects of MPC supplementation in combination with RT on the health aspects of RT practitioners.

#### 4.2 Characteristics of the MPC

The low fat values of the protein concentrate suggest the effectiveness of skimmed milk used as raw material, which can later be used in the production of ricotta from skimmed milk (Table 3). The percentage of fat in whole and skimmed milk was in accordance with IN 76 of 2018, which suggests that whole milk must have a fat content greater than 3.0% and skimmed milk must have a fat content below 0.5% fat (Brasil, 2018). Regarding the protein analysis, the results showed that the MPC was in compliance with Brazilian legislation (Brasil, 2018), with 3.17% of protein in whole milk and 3.19% of protein in skimmed milk.

According to Doska et al. (2012) casein is the most predominant protein of dairy proteins, in the results of milk analyses, differences were observed between the types of milk; however, both milk types were in compliance with the Brazilian legislation [minimum content of lactose: of 4.3, Brasil (2018)]. The TDE results of the present study were similar to those obtained by Brasil et al. (2012). The average values of DDE

**Table 2.** Comparison of groups (control vs. intervention) regarding the differences between post-test and pre-test (delta) for body composition variables.

Variables	CG (n = 15) Mean ± SD	IG (n = 15) Mean ± SD	p-value
Delta Body weight	0.22 ± 1.89	2.08 ± 3.14	0.06
Delta BMI	0.11 ± 0.63	0.53 ± 1.22	0.25
Delta Fat %	-2.30 ± 1.16	-6.08 ± 1.57	< 0.01*
Delta Lean mass	2.12 ± 1.30	6.40 ± 2.18	< 0.01*
Delta Fat mass	-1.91 ± 1.40	-4.57 ± 1.45	< 0.01*

BMI: body mass index; SD: standard deviation. \*p-value < 0.01 according to the t test for independent samples or the Mann Whitney U test.

in whole and skimmed milk were also in accordance with IN 76 (Brasil, 2018) which suggests milk must contain a minimum value of 8.40% defatted solids.

In relation to SCC, skimming milk for ricotta production greatly reduced the percentage of SCC/mL. (Table 3). Santos et al. (2007) also reported that skim-associated microfiltration was effective in removing somatic cells from milk, with reductions of 92.5% and 99.5% for low- and high SCC milk, respectively. However, in the present study, the mean SCC values of whole and skimmed milk were lower than those reported by Silva et al. (2009). It is noteworthy that the milk used in this study was in accordance with the legislation in force in Brazil (Brasil, 2018), which recommends a maximum value of 500,000 SCC/mL for receiving milk. The mean TBC values in the present study are in accordance with IN 76, which recommends a maximum bacterial count of 300,000 CFU per mL of milk (Brasil, 2018).

The percentage of fat in whole milk yogurt, skimmed milk ricotta, and MPC was higher than that reported by Antunes et al. (2015), with a value of 2.70%. The value of the protein supplement of milk concentrate was higher than that reported by Batista et al.

**Table 3.** Mean values and standard deviation of fat, protein, casein, lactose, total dry extract (TDE), defatted dry extract (DDE), somatic cell count (SCC), log SCC, total bacterial count (TBC) and log TBC of milk (whole and skimmed) used in the processing of natural whole yogurt and skimmed milk ricotta.

Variables	Milk		CV (%)
	Whole	Skimmed	
Fat (%)	3.26 ± 0.31 <sup>a</sup>	0.31 ± 0.13 <sup>b</sup>	13.11
Protein (%)	3.17 ± 0.12 <sup>a</sup>	3.19 ± 0.16 <sup>a</sup>	4.53
Casein (%)	2.43 ± 0.13 <sup>b</sup>	2.51 ± 0.17 <sup>a</sup>	6.08
Lactose (%)	4.40 ± 0.08 <sup>b</sup>	4.55 ± 0.06 <sup>a</sup>	1.66
TDE (%)	11.70 ± 0.39 <sup>a</sup>	8.98 ± 0.24 <sup>b</sup>	3.13
DDE (%)	8.45 ± 0.21 <sup>b</sup>	8.66 ± 0.22 <sup>a</sup>	2.57
SCC (SC/mL)	200583 ± 69100 <sup>a</sup>	59167 ± 17875 <sup>b</sup>	38.86
Log of SCC	5.28 ± 0.13 <sup>a</sup>	4.74 ± 0.17 <sup>b</sup>	3.03
TBC (CFU/mL)	7375 ± 5026 <sup>a</sup>	8167 ± 2319 <sup>a</sup>	50.37
Log of TBC	3.78 ± 0.28 <sup>b</sup>	3.89 ± 0.15 <sup>a</sup>	5.87

CV: coefficient of variation. CFU: Colony Forming Unit. Different letters on the same line differ from each other ( $p < 0.05$ ) using Tukey's test at 5% significance.

(2015), the authors reported an average of 8.63% of protein in the product developed with the addition of 37.0% of WP, a lower value was also reported by Antunes et al. (2015), who reported an average of 6.18% protein in semi-skimmed yogurt added to 35.0% WP.

The average amount of carbohydrates was significantly different between the products, being higher in yogurt and lower in ricotta. The percentage of carbohydrates in this study was lower than that reported by Bezerra et al. (2019), who evaluated the carbohydrate values, physical-chemical, and sensory profile of natural Greek yogurts made with different concentrations of sucrose.

The ash and dry matter content also differed significantly between the products (Table 4). The results of the mineral matter in this study were higher than those reported by Martins et al. (2013) when evaluating yogurt made with water-soluble soybean extract supplemented with inulin, the average ash value was 0.48%. The values were also higher than that reported by Bezerra et al. (2019), who evaluated the ash content in natural Greek yogurts made with different sucrose concentrations was 0.89% ash. The mean dry matter values in this study were higher than those reported by Antunes et al. (2015), who reported an average value of 22.24%. The water content reported by Antunes et al. (2015) was superior to the present experiment with an average water content was 83.29% in traditional semi-skimmed yogurt and 77.76% in the semi-skimmed yogurt supplemented with 35% of WP.

The calorific value (kcal) of the evaluated products were also different (Table 4). Calorific values lower than those reported in this study were reported by Santana et al. (2015) when analyzing the physicochemical and nutritional profile of pitaya-based yogurt (*Hylocereus undatus*) enriched with quinoa (*Chenopodium quinoa*) and sucralose. However, the protein concentration in the aforementioned study was 5.0%, which is lower than that in this report. It is worth mentioning that for RT practitioners and the people, who seek a healthy diet with a low concentration of carbohydrates, this product can meet these needs. According to Haraguchi et al. (2006), RT practitioners have protein demands of 1.6 to 1.7 g per kg (body weight) per day. Thus, the caloric value of our MPC will possibly help muscle recovery and development, since the MPC

**Table 4.** Mean values and standard deviation of fat, protein, carbohydrate, ash, dry matter (DM), water content, caloric value and pH of natural whole milk yogurt, skimmed milk ricotta and MPC.

Variables	Products		
	Natural whole milk yogurt	Skimmed milk ricotta	MPC
Fat (%)	3.13 ± 0.28 <sup>a</sup>	2.90 ± 0.07 <sup>b</sup>	3.32 ± 0.08 <sup>a</sup>
Protein (%)	3.08 ± 0.04 <sup>c</sup>	23.33 ± 0.42 <sup>a</sup>	18.51 ± 0.51 <sup>b</sup>
Carbohydrate (%)	12.42 ± 0.69 <sup>a</sup>	0.98 ± 0.81 <sup>c</sup>	4.58 ± 0.59 <sup>b</sup>
Ashes (%)	0.99 ± 0.02 <sup>c</sup>	2.17 ± 0.04 <sup>a</sup>	1.86 ± 0.04 <sup>b</sup>
DM (%)	19.62 ± 0.79 <sup>c</sup>	29.40 ± 0.64 <sup>a</sup>	26.41 ± 0.46 <sup>b</sup>
Water content (%)	80.38 ± 0.79 <sup>a</sup>	70.59 ± 0.64 <sup>c</sup>	73.59 ± 0.46 <sup>b</sup>
Caloric Value (kcal)	90.21 ± 3.99 <sup>b</sup>	123.53 ± 2.55 <sup>a</sup>	122.26 ± 1.85 <sup>a</sup>
Ph	4.12 ± 0.05 <sup>c</sup>	5.32 ± 0.05 <sup>a</sup>	5.22 ± 0.05 <sup>b</sup>

Different letters on the same line differ from each other ( $p < 0.05$ ) using Tukey's test at 5% significance.

**Table 5.** Mean results and acceptance index of appearance, color, taste, aroma, texture and global evaluation of MPC for RT practitioners.

Attributes	Groups				CV(%)
	CG (n = 15)	AI (%)	IG (n = 15)	AI (%)*	
Appearance	6.40 ± 0.91 <sup>a</sup>	75.00	6.66 ± 0.62 <sup>a</sup>	75.00	11.90
Color	6.47 ± 0.52 <sup>a</sup>	85.71	6.53 ± 0.52 <sup>a</sup>	75.00	7.94
Taste	6.20 ± 0.68 <sup>b</sup>	85.71	6.66 ± 0.59 <sup>a</sup>	87.50	9.16
Aroma	6.80 ± 0.68 <sup>a</sup>	87.50	6.93 ± 0.59 <sup>a</sup>	87.50	9.27
Texture	6.13 ± 0.74 <sup>a</sup>	71.43	6.20 ± 0.88 <sup>a</sup>	87.50	12.31
Global evaluation	6.46 ± 0.64 <sup>b</sup>	85.00	7.26 ± 0.46 <sup>a</sup>	87.50	10.54
Purchase intention	3.66 ± 0.41 <sup>b</sup>	N/A	4.33 ± 0.37 <sup>a</sup>	NM	13.91

CV: coefficient of variation. AI: acceptability index. NM: not measured. Different letters on the same line differ from each other ( $p < 0.05$ ) using Tukey's test at 5% significance.

has 18.51% of proteins. The average pH of milk ricotta was slightly higher than that of MPC, while yogurt had a pH lower than 5 (Table 4). Mean pH lower than that in the present study was reported by Bezerra et al. (2019), who analyzed the pH of natural Greek yogurts made with different concentrations of sucrose, reporting average pH values between 4.25 and 4.51. What justifies the higher average value in our study was the addition of milk ricotta, since it is considered cheese, and the pH of the cheese was higher than that of yogurt.

#### 4.3 Sensory profile of MPC

In the sensory evaluation of the milk-based protein supplement for RT practitioners (Table 5), both groups rated the milk-based protein supplement on the hedonic scale between “I liked it slightly” and “I liked it moderately”. The scores for these attributes may have been influenced by the evaluation because the MPC supplement had some visible and perceptible lumps during ingestion, which suggests the importance of further exploration in future research. The flavor attribute differed significantly between the groups, being classified in the hedonic scale as “I liked it slightly” and “I liked it moderately”, and in the CG it was classified as “I liked it slightly”. This difference between the groups may have occurred because the perception of flavor is subjective and influenced by previous experiences and cultural factors. When comparing the global assessment attribute, the mean value also differed between the groups, with the IG rating the protein supplement as moderately liked, while the control group rated it as slightly liked.

As for the acceptability of the product, even with no difference in the averages obtained for appearance, color, and aroma, the acceptance of both groups was good, not showing disapproval by the evaluators. The MPC when evaluated for the purchase intention was different between groups, perhaps “buying/maybe not buying” from the CG and “possibly buying” from the IG, demonstrating that the evaluators could possibly buy the product if it was available in supermarkets. The percentages of the acceptability index of the milk protein concentrate were above 71% for all attributes in both groups. The results of the sensory analysis in this study indicated that an acceptability index above 70% for the attributes of appearance, aroma, flavor, and texture of a product indicates viability for commercial investment (Costa et al., 2013).

The flavor with the highest preference reported by the CG was chocolate, followed by strawberry, which may have been influenced by the fact that most yogurts and dairy drinks in the market have these flavors. On the other hand, the results of the sensory analysis regarding the suggestions for the product may have been influenced by the lack of flavor or sugar in the MPC. The participants of the IG reported feeling “strength gain,” “feeling of less fat,” “muscle mass gain,” “feeling of a rested body,” “firm muscles” and “more disposition”. The “feeling less body fat,” was supported by a decrease in fat mass during the consumption of MPC and an increase in lean mass (Table 5).

To better understand the sensory aspects of the product, it is necessary to repeat the experiment with a larger number of volunteers.

## 5 Conclusion

MPC with low fat and carbohydrates and high protein concentration directly influenced the decrease in the percentage of fat, fat mass, and lean mass increase in RT practitioners. In addition, the lipid profile evaluated in RT practitioners was not altered in either the CG or IG, demonstrating that the product consumed by the volunteers did not seem to present possible health problems.

The processing of skimmed whole milk was efficient, and the whole and skimmed milk used for processing the MPC were within the standards of physical-chemical parameters, SCC, and TBC established by Brazilian legislation. The sensory analysis showed that the MPC had good appearance, color, aroma, texture, global evaluation, and acceptability index. Due to the high acceptance index, the report suggests that the product could have potential to be introduced in the market for RT practitioners.

However, further studies should be carried out to improve MPC in terms of shelf life, new formulations, and better analysis of the impacts on performance and health.

## Conflict of interest

The authors declare no conflict of interest.

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