Original Article

Compressed food with added functional oligopeptides improves performance during military endurance training

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Background and Objectives: Oligosaccharide or oligopeptide supplementation may have a significant impact on endurance performance. This study evaluated the effects of adding maltooligosaccharides (MO) or soy oligopeptides (SO) to compressed food (CF) on the physical response of soldiers to daily military training. Methods and Study Design: Twelve soldiers were randomized to four diet groups: regular meals, CF, CFMO, and CFSO (crossover design). They participated in exercise tests including 90 minutes running at 55-65% VO₂max and exhaustive running. Heart rates, rating of perceived exertion (RPE), and blood and urine samples were collected during exercise and recovery. Results: The recovery heart rates were significantly lower with the CFMO diet compared with the other diets. Compared with all other diets, blood glucose levels were higher, post-exercise blood lactate levels were lower, and lactate clearance during recovery was higher with the CFMO diet, followed by the CFSO diet. Post-exercise levels of erythrocytes and hematocrit were significantly higher with the CFSO diet. Post-exercise urine specific gravity was lower with the CFMO diet and urine pH was decreased with the CFSO diet. Blood urea nitrogen (BUN) and uric acid (UA) were significantly higher with the CFSO diet than with the other diets. There was no significant difference in skeletal and cardiac muscle injury indices and RPE among diets. Conclusions: CFMO led to better heart rate recovery, improved and maintained blood glucose and increased removal of blood lactate. CFSO accelerated removal of blood lactate during recovery, maintained oxygen supply, and increased fluid retention.

Key Words: compressed food, oligosaccharide, oligopeptide, exercise performance, endurance training

INTRODUCTION

Military personnel on the battlefield are usually provided with ready-to-eat or ready-to-cook packed foods that have specific characteristics like storage stability with long shelf life, transport convenience, nutritional value, and easy preparation. Use of thermally processed canned or compressed food (CF) that can be consumed with or without prior heating, are extensively used in military nutrition.^{1,2} CF is the major source of food-derived energy in the Chinese military. However, the food intake is inadequate when packaged military rations are used as the sole source of food, which can lead to health problems like nutritional deficiency and hence reduce vigor and performance.³ There is a need for improvement in the quality and composition of CF to enhance its nutritional and functional values. Foods with adequate nutritional value would allow military personnel delivering optimal performance during deployment.

Oligosaccharides and oligopeptides are food materials with high nutritional value that are supposed to improve athletic performance. It was observed that addition of protein to a carbohydrate supplement enhanced aerobic endurance performance.⁴ Studies showed that fructooligosaccharides increased the absorption of calcium, magnesium, and iron, and decreased the occurrence of osteoporosis.⁵ Another study showed that oligopeptides extracted from hen's egg yolks could potentially suppress the development of hypertension in spontaneously hypertensive rats, possibly by the inhibition of the angiotensinconverting enzyme (ACE).⁶ Oligopeptides derived from soybean can be used as natural antioxidants or antihypertensive compounds in the pharmaceutical and functional food industries.⁷ Soy oligopeptides (SO) can be effective in pain control by increasing the absorption of opioids compared with a control mixture of proteins in healthy adult men.⁸

The functional characteristics of oligosaccharides and oligopeptides have been studied extensively, but few studies have been performed on human volunteers to examine the effects of oligosaccharides and oligopeptides to improve the sports ability and to improve the fatigue re-

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covery after exercise. In addition, many exercise and endurance training studies used parameters like blood lactate, heart and lung functions, blood and urine biochemistry, detection of muscle injury markers, and subjective ratings of perceived exertion (RPE), because these parameters are most likely to reflect muscle activity, breathing, and metabolic changes during and after exercise.^{9,10} Therefore, the aim of the present study was to use these parameters to evaluate the effects of maltooligosaccharides (MO)- and SO-enriched CF on exercise performance and endurance training in military personnel.

MATERIALS AND METHODS

Test samples

The CF, compressed food with maltooligosaccharides (CFMO), and compressed food with soy oligopeptide (CFSO) were developed by the Quartermaster Institute of the General Logistic Department of the Chinese People's Liberation Army (CPLA), China.

The CF was made of biscuit powder (83%), vegetable oil (11%), vitamins (0.3%), minerals (0.7%), and water (5%). The biscuit formula included wheat flour (55%), white granulated sugar (13.5%), salt (0.5%), vegetable oil (13.5%), milk powder (3%), baking soda (0.2%), and water (14.3%). The CF was made in accordance with the above formula using the CF Technology (Patent: No. ZL 200710097841.9 [China]) including two parts of the biscuit making and biscuits crushed briquetting. Taurine, carnitine, caffeine, and any other banned substances were not added. The CF provided 1880 kJ/100 g, of which 13% were from protein, 30% from fat, and 57% from carbohydrates. Each 100 g of CF contained 14.7 g of protein, 15.2 g of fat, and 63.9 g of carbohydrates.

The CFMO was made by adding 5% of MO to the biscuit dough during the late stages of CF processing. MO is a maltose compound that consists of glucose (1.0-4.0%), maltose (18.0-23.0%), malto-triose (20.0-25.0%), maltotetraose (8.0-13.0%), malto-pentaose (4.0-8.0%), maltohexaose (3.0-7.0%), and malto-heptaose and other carbohydrates (20.0-46.0%).

The CFSO was made by adding 5% of SO to the biscuit dough during the late stages of CF processing. SO is a mixture of small peptides; 90% of these peptides have a molecular weight <1000 Da.

Study participants

Sixty soldiers were screened from a military base in Beijing; 34 soldiers were selected based on their fitness, as determined by their resting heart rates, recent 5-km running records, and training performance. After blood examination and fitness, 12 soldiers were selected for this trial.

Inclusion criteria: (1) 1-1.5 years of service; and (2) able to participate in standard military training. Exclusion criteria: (1) heart rate of <60 or >100 bpm at rest or >180 bpm after a 2400-m run in 12 mins; (2) unsuccessful fitness test; (3) unstable overall level of training; (4)strong subjective feeling of fatigue; (5) failed treadmill fitness test; (6) white blood cell (WBC) count outside the normal range ((4.0-10.0)×10⁹/L); (7) overweight (BMI >23.9 kg/m²) or underweight (BMI <18.5 kg/m²); or (8) substandard appetite. Soldiers were randomized to four groups using the drawing lots method (n=3/group).

The study was approved by the Ethics and Research Committee of the Quartermaster Institute of the General Logistic Department of CPLA and was conducted in accordance with the Declaration of Helsinki. All participants provided a written informed consent before enrollment.

Study design

This was a crossover design study. All participants were successively submitted to all four diets. The four food types were regular meal, CF, CFMO, and CFSO. Each round lasted for 7 days, including 4 days for the consumption of the food followed by a 3-day washout during which normal diet was allowed (Table 1).

During the 4-day experimental period, soldiers had to strictly adhere to the consumption of the experimental food (3300 kcal/day). During the 3-day normal diet period, there was no food restriction, but snacks were not allowed between meals and the daily intake of calories had to be <4000 kcal. All other kinds of foods, supplements, cigarettes, and alcohol were prohibited during the trial.

Study procedure

The following procedures were implemented during each testing round:

Days 1 and 2: Diet according to the pre-set experimental schedule; all participants were submitted to the same amount and intensity of daily training. Day 3: Diet according to the pre-set experimental schedule; all participants participated in simple activities, without high intensity workout. Day 4: Diet according to the pre-set experimental schedule; all participants were submitted to exercise capacity test, as explained below.

(1) The participants provided fasted midstream urine in

Test round	Days	Regular meal	CF	CFMO	CFSO	
1st round	Day 1-4 Day 5-7	A ₁ -A ₃ Free diet	B ₁ -B ₃	C ₁ -C ₃	D ₁ -D ₃	
2nd round	Day 1-4 Day 5-7	D ₁ -D ₃ Free diet	A ₁ -A ₃	B_1 - B_3	C ₁ -C ₃	
3rd round	Day 1-4 Day 5-7	C ₁ -C ₃ Free diet	D ₁ -D ₃	A ₁ -A ₃	B ₁ -B ₃	
4th round	Day 1-4 Day 5-7	B ₁ -B ₃ Free diet	C ₁ -C ₃	D ₁ -D ₃	A_1 - A_3	

 Table 1. Crossover experiment design

CF: compressed food; CFMO: compressed food with maltooligosaccahrides; CFSO: compressed food with soy oligopeptides.

the morning and venous fasted blood. Blood sugar and lactate were measured. (2) Diet according to the pre-set experimental schedule, followed by a 30-min break; blood sugar and lactate were measured. (3) Running for 90 minutes at 55-65% VO2max. Heart rate was monitored and urine samples were obtained immediately afterwards. Blood sugar and lactate were measured every 45 mins. Blood lactate was measured again after resting for 10 min. (4) Exhaustive running test starting at 8 km/h, increasing to 14 km/h steadily within 3 minutes (equivalent to 85-92% VO₂max), and maintained at 14 km/h until exhaustion. Heart rate was monitored. Blood lactate was measured immediately and 3, 9, and 15 min after exhaustion. (5) On Day 5, fasted venous blood and midstream urine were obtained in the morning for biochemical marker measurement. All blood samples to determine blood sugar and lactate levels were collected from fingertips.

Daily training

Daily training included 5 000-m running, 400-m barrierrunning, sit-ups, push-ups, fighting movements, grenade throwing, blasting, and shelter construction.

Testing indices and methods

Heart and lung functions were determined using Cortex MetaMax cardiopulmonary function 3B telemetry. Blood glucose was measured using a One Touch Ultra blood glucose system. Blood lactate was determined using a YSI 1500 lactate analyzer. Blood lactic acid value immediately after exhaustive running was taken as reference. Blood lactate clearance rate (%) = [(reference value blood lactic acid value 3/9/15 min after exhaustive running) / reference value)] * 100%. For routine blood tests (WBC, red blood cell, hemoglobin (HGB), hematocrit (HCT), platelet (PLT)), a Siemens ADVIAR 2120 whole blood cell analyzer was used. For routine urine analysis (specific gravity and pH), a Bayer Clinitek Status urine analyzer was used. Assessment of skeletal and cardiac muscle injury (creatinine kinase (CK), myoglobin, cardiac troponin I (TnI)), liver function (alanine aminotransferase (ALT), aspartate aminotransferase (AST), total protein (TP), albumin (ALB)), and kidney function (creatinine (CRE), blood urea nitrogen (BUN), uric acid (UA)) indexes were measured using a Beckman Coulter Unicel DXC600 automatic biochemical analyzer and Beckman Coulter UnicelTM DXI800 automatic light-emitting device.

To assess subjective feeling of fatigue, the Borg's perceived exertion and pain scale ratings of perceived exertion (RPE) were used.¹¹ The subjective perception of fatigue during each stage of exercise was scored by the participants on a scale ranging from 6 to 20, where 6-8 was considered very easy, 9-10 fairly easy, 11-12 easy, 13-14 somewhat tired, 15-16 tired, 17-18 very tired, and 19-20 exhausted. During the whole trial, each value of the Borg's 6-20 category scale was explained repeatedly to participants. After running for 10-15 min, 30-35 min, 50-55 min, 70-75 min, 85-90 min, and at the end of exhausted running, the experimenters held up the scale and the participants reported their RPE at that instant, which was used to estimate total body exhaustion at these time points.

Statistical analysis

Statistical analyses were performed using SPSS 13.0 (SPSS Inc., Chicago, IL, USA). The data were presented as mean±standard error (SE) and analyzed using one-way analysis of variance (ANOVA) with the least-significant difference (LSD) method for multiple comparisons or the Tamhane's T2 method if variables were non-normally distributed. A dynamic curve of different monitoring time points was analyzed by the general linear model via comparing univariate curve slopes and intercept differences among groups. Differences were considered to be significant when p<0.05.

RESULTS

Demographic and other baseline characteristics

Participants were 19.8 \pm 0.5 years of age; their height was 171.9 \pm 1.3 cm, body weight was 65.2 \pm 1.8 kg, and body mass index (BMI) was 22.0 \pm 0.4 kg/m². The maximum oxygen uptake (VO_{2max}) was 57.9 \pm 0.6 mL/min/kg. No significant difference was observed among the four groups.

Changes in heart rate during exercise

Heart rate (HR) during the 55-65% VO₂max running exercise for 90 min showed that participants who took CF (CF, CFSO, or CFMO) had higher HR compared with participants who took normal diet (Figure 1A). In addition, there were significant differences in the 85-92% VO₂max with exhaustive running test between the regular diet and the CFMO, CF, and CFSO diets (all p<0.01 vs. the regular diet) (Figure 1B). HR after exhaustive running was lower with the CFMO diet compared with the CF and CFSO diets at each time point (all p<0.01) (Figure 1C).

Changes in blood glucose during exercise

There was no significant difference in morning fasting blood glucose among all diets. Thirty minutes after food consumption, blood glucose was higher with the CFMO diet compared with the regular diet, CF, and CFSO. Blood glucose was lower with all diets after 55-65% VO₂max running for 90 min. The blood glucose was higher with the CFMO diet compared with the others (all p<0.05) (Figure 2).

Changes in blood lactate during exercise

Blood lactate levels increased rapidly with all diets during 55-65% VO2max running for 45 min. During 85-92% VO2max exhausted running, blood lactate increased rapidly to a maximum value with all diets. During the 15min recovery, blood lactate decreased with all diets. Blood lactate was the lowest with the CFMO diet at all time points, while they were the highest with the regular diet. A significant difference was observed for blood lactate among the CFMO, CFSO, CF, and regular diets (p < 0.01) (Figure 3A). Blood lactate after the 10-min relaxation period was the lowest with the CFMO diet, followed by the CFSO, CF, and regular diets (p < 0.01) (Figure 3A). The blood lactate clearance was the fastest with the CFMO diet, followed by the CFSO diet, but there was no significant difference among the four diets after exhaustive running (Figure 3B).



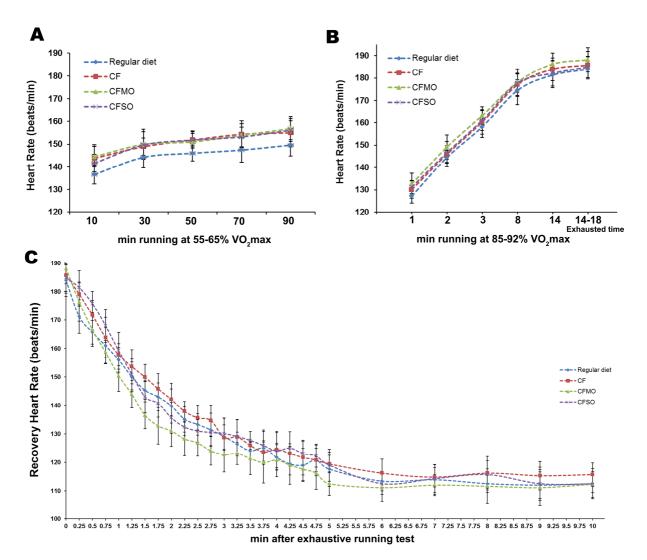


Figure 1. Heart rate differences with different compressed food consumption. (A) Changes in heart rate during 55-65% VO₂max running for 90min testing. (B) Changes in heart rate during 85-92% VO₂max running until exhaust. (C) Changes in heart rate after exhaustive running. The data are shown as mean±standard error (SE). CF: compressed food, CFMO: compressed food with maltooligosaccahrides, CFSO: compressed food with soy oligopeptides.

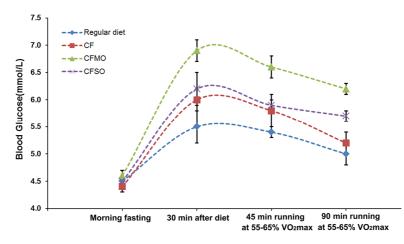


Figure 2. Changes of blood glucose concentrationduring exercise with different compressed food consumption. The data are shown as mean±SE.

Changes in routine blood parameters

No significant difference was observed in RBC count, platelet count, or hemoglobin at any time point among all four diets (all p>0.05) after food consumption for four consecutive days. Compared with the other diets, RBC count was significantly higher with the CFSO diet imme-

diately post-exercise (p<0.05). Morning fasting hematocrit values were significantly higher with the CFSO diet compared with the regular diet (p<0.05) and significantly higher with the CFMO and CFSO diets compared with the regular diet immediately after exercise (p<0.05) (Table 2).

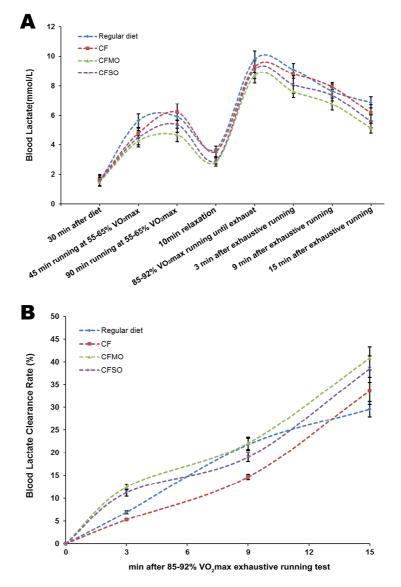


Figure 3. Blood lactate differences with different compressed food consumption. (A) Changes in blood lactate concentration during exercise. (B) Changes in blood lactate clearance rate (%) after exhaustive running. The data are shown as mean±SE.

Changes in urine specific gravity and urine pH

The urine specific gravity with the CFMO diet was lower compared with the CF and CFSO diets after 90-min running (p<0.05). In addition, the increase in urine specific gravity was smaller with the CFMO diet compared with the other diets (p<0.05). A decrease in urine pH was observed with the CFSO diet immediately after exercise (p<0.05) (Table 3).

Changes in liver or renal function parameters

After four days of food consumption, the liver function indexes of all participants were within the normal range and were not significantly different from each other (Table 2). There was no significant difference for serum creatinine levels. The BUN with the CFSO diet was significantly higher than with the other diets after fasting, immediately after exercise, and the next morning after exercise (p<0.01, 0.01, 0.05 or 0.01). The lowest BUN was observed at each monitoring time point with the CF diet. The BUN was significantly lower with the CF diet compared with the regular diet (p<0.05) immediately after exercise and the next morning after exercise. UA after food consumption with all diets was within the normal range, but the UA levels with the CFSO diet were significantly higher compared with the other diets (p<0.05 or 0.01) (Table 3).

Changes in indices of skeletal or cardiac muscle injury

Table 4 presents the changes in skeletal and cardiac muscle injury indices. No significant difference was observed in myoglobin, CK, or troponin at any time points. After exhaustive exercise, the highest myoglobin levels were observed with the CFSO diet, followed by the CFMO and CF diets, compared with the regular diet, but the differences were not statistically significant. After exhaustive exercise, CK was the lowest with the CFMO diet and the highest with the regular diet, but no significant difference was observed between these diets. In addition, no significant difference was observed for troponin among all diets after exercise.

RPE scores during exercise performance test

Table 5 presents the RPE scores of the soldiers during exercise. The RPE scores increased continuously during

Diets	Time point	White blood cell (×10 ⁹ cells/L)	Red blood cell /Erythrocyte (×10 ¹² cells/L)	Hemoglobin (g/L)	Hematocrit (%)	Platelet (×10 ⁹ cells/L)	Alanine aminotransferase (U/L)	Aspartate aminotransferase (U/L)	Total protein (g/L)	Albumin (g/L)
Baseline (n=12)	Morning fasting	5.9±0.49	4.60±0.05	147±1.6	40.7±0.4	230±15	27.1±2.1	23.6±1.1	75.5±1.0	45.5±0.6
Regular meal	Morning fasting	5.9±0.39	4.57±0.07	146±2.2	39.1±0.6	221±12	27.3±2.2	23.8±1.4	79.7±0.9	47.5±0.6
(n=12)	Immediately after exercise	10.4 ± 0.48	4.75±0.06	152±1.9	40.4 ± 0.4	285±14	42.1±2.6	32.4±1.9	88.6±0.9	52.5±0.5
	The next morning after exercise	6.0±0.33	4.50±0.05	143±2.2	38.4±0.6	226±12	28.6±2.6	27.6±3.1	79.5±1.5	48.0±0.6
CF	Morning fasting	6.0±0.35	4.68±0.07	149±2.1	40.1±0.6	235±11	26.5±2.2	22.0±1.0	81.0±0.9	48.3±0.4
(n=12)	Immediately after exercise	9.3±0.60	4.84 ± 0.08	156±2.5	41.6±0.7	303±16	42.6±4.0	30.2±1.9	93.5±1.6	53.4±0.7
	The next morning after exercise	6.1±0.31	4.60±0.05	146±1.4	39.2±0.3	240±12	26.3±2.1	23.3±1.5	78.4±1.2	47.4±0.6
CFMO	Morning fasting	6.0±0.32	4.67±0.05	149±1.4	40.0±0.3	230±10	25.7±2.0	22.1±0.9	79.4±0.5	47.4±0.4
(n=12)	Immediately after exercise	10.2±0.44	4.89±0.06	157±2.2	$42.2\pm0.5^{\dagger}$	287±13	39.9±2.7	30.2±1.0	90.7±1.2	52.3±0.6
()	The next morning after exercise	6.0±0.34	4.55±0.06	144±1.9	39.0±0.5	229±10	26.8±1.9	24.9±1.1	78.2±0.7	46.9±0.5
CFSO	Morning fasting	6.1±0.33	4.73±0.08	152 ± 2.0	$40.8 \pm 0.6^{\dagger}$	216±8	28.6±3.2	24.3±1.0	82.0±1.0	48.7±0.4
(n=12)	Immediately after exercise	10.3±0.46	$4.96{\pm}0.08^{\dagger}$	159±2.3	$42.3 \pm 0.7^{\dagger}$	287±13	42.8±3.5	33.5±1.1	94.1±1.1	54.1±0.8
· · ·	The next morning after exercise	6. 6±0.50	4.60±0.07	147±2.4	39.3±0.6	225±12	31.4±3.4	31.0±4.4	81.5±1.4	48.6±0.7

Table 2. Changes in routine blood parameters and liver function

CF: compressed food; CFMO: compressed food with maltooligosaccharides; CFSO: compressed food with soy oligopeptides.

Data are shown as mean±SE. $^{\dagger}p<0.05$ vs Regular meal at the same time point.

Diets	Time point	Urine specif- ic gravity	pН	Creatinine (ummol/L)	Blood urea nitrogen (mmol/L)	Uric acid (mg/dL)
Baseline (n=12)	Morning fasting	1.03±0.001	6.13±0.09	85±2.0	6.60±0.29	5.68±0.19
Regular meal	Morning fasting After running for 90 min	1.03±0.001 1.02±0.002	6.14±0.10 6.21±0.19	79±1.9	6.38±0.23	5.50±0.18
(n=12)	Immediately after exercise The next morning after exercise	1.03±0.002 1.03±0.001	5.86±0.12 6.14±0.10	102±4.8 79±2.4	7.61±0.24 7.46±0.18	5.97±0.21 5.64±0.18
CF (n=12)	Morning fasting After running for 90 min	1.03±0.001 1.02±0.002	6.08±0.06 6.27±0.25	85±2.7	6.03±0.27	5.40±0.20
` ,	Immediately after exercise The next morning after exercise	1.03±0.000 1.03±0.001	5.80±0.08 6.13±0.15	102±5.2 81±2.3	6.75±0.31 [†] 6.53±0.29 [†]	5.78±0.28 5.66±0.17
CFMO (n=12)	Morning fasting After running for 90 min	1.03±0.002 1.02±0.002 [‡]	6.50±0.14 6.25±0.14	83±2.2	6.37±0.19	5.26±0.17
	Immediately after exercise The next morning after exercise	1.03±0.001 [‡] 1.03±0.002	6.00±0.01 6.25±0.10	100±4.3 86±1.9	7.43±0.20 6.91±0.29	5.66±0.25 5.55±0.20
CFSO (n=12)	Morning fasting After running for 90 min	1.03±0.001 1.03±0.002 [§]	6.00±0.01 5.96±0.13	85±2.5	7.68±0.24 ^{++‡‡§§}	6.26±0.26 ^{†‡‡§§}
. ,	Immediately after exercise The next morning after exercise	1.03±0.001 ^{†§} 1.03±0.001	5.70±0.08 [§] 6.13±0.13	103±5.4 83±3.1	9.23±0.29 ^{††‡‡§§} 8.67±0.35 ^{†‡‡§§}	6.79±0.34 ^{†‡§§} 6.42±0.30 ^{†‡§§}

Table 3. Changes in routine urine parameters and in renal function

CF: compressed food; CFMO: compressed food with maltooligosaccharides; CFSO: compressed food with soy oligopeptides. Data are shown as mean±SE.

 $p^{*} = 0.05$, $p^{*} = 0.01$ vs Regular meal; $p^{*} = 0.05$, $p^{*} = 0.01$ vs CF; $p^{*} = 0.05$, $p^{*} = 0.01$ vs CFMO at the same time point.

Table 4. Changes in skeletal and cardiac muscle injury indi	ces
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Diet	Time point	Creatine kinase (U/L)	Myoglobin (ng/mL)	Cardiac troponin I (ng/mL)
Baseline (n=12)	Morning fasting	216±39.1	22.4±1.6	0.021±0.003
Regular meal (n=12)	Morning fasting Immediately after exercise The next morning after exercise	175±30.8 236±40.2 218±33.2	20.1±1.3 63.8±25.3 27.6±4.5	$\begin{array}{c} 0.014{\pm}0.002\\ 0.016{\pm}0.002\\ 0.021{\pm}0.003\end{array}$
CF (n=12)	Morning fasting Immediately after exercise The next morning after exercise	163±18.1 223±26.2 226±42.3	22.1±1.4 49.5±11.2 24.5±2.9	0.015±0.002 0.020±0.005 0.020±0.008
CFMO (n=12)	Morning fasting Immediately after exercise The next morning after exercise	145±13.7 206±17.4 266±30.9	20.2±1.2 51.7±11.7 26.2±1.8	$\begin{array}{c} 0.014{\pm}0.003\\ 0.022{\pm}0.004\\ 0.026{\pm}0.006\end{array}$
CFSO (n=12)	Morning fasting Immediately after exercise The next morning after exercise	149±8.7 209±10.9 210±33.5	21.7±1.2 77.3±27.5 41.0±12.6	0.016±0.003 0.018±0.002 0.021±0.002

CF: compressed food; CFMO: compressed food with maltooligosaccharides; CFSO: compressed food with soy oligopeptides. Data are shown as mean±SE.

55-65% VO_{2max} 90-min running, but there was no significant difference among the diets (Table 5).

DISCUSSION

CF is an important part of military rations and nutrition, but food intake is inadequate when packaged military rations are used as the sole source of food, eventually leading to problems like nutritional inadequacy and hence reducing vigor and performance. The aim of this study was to evaluate the effects of MO- and SO-enriched CF on exercise performance and endurance training in military personnel.

HR during exercise measures cardiac load and HR recovery, and may reflect the state of the autonomic nervous system and the body's capacity to respond to exercise.¹² The present study suggested that the CFMO diet could accelerate HR recovery after daily training. The amount of muscle glycogen stores, which is the main fuel for muscular work during prolonged high-intensity exercise, is an important factor in determining endurance capacity. Supplementation with carbohydrates increases endurance performance and enhances lactate removal, and thereby delays the onset of fatigue.¹³

Blood glucose is an indicator of fatigue and athletic performance. Carbohydrates are the main source of blood glucose. Carbohydrate ingestion during exercise may elevate blood glucose and increase endurance capacity.¹⁴ The present study showed higher blood glucose levels with the CFMO diet compared with the other diets, which could be possibly because of added MO. Based on these results, an inference can be drawn supporting the CFMO diet over the other study diets for improving and maintaining blood glucose during longer and moderate exercises.

		Exhausted				
Diet	Running for	running				
	10-15 min	30-35 min	50-55 min	70-75 min	85-90 min	Tunning
Regular meal (n=12)	7.8±0.6	11.3±0.6	13.6±0.6	14.7±0.7	16.8±0.8	19.0±0.8
CF (n=12)	8.2±0.5	11.0±0.6	13.4±0.8	14.1±0.8	15.6±0.9	18.7±0.9
CFMO (n=12)	8.1±0.7	11.3±0.7	13.7±0.8	15.3±0.9	16.5±0.8	18.3±0.4
CFSO (n=12)	8.0±0.4	11.7±0.5	13.7±0.7	14.7±0.8	16.6±0.8	19.1±0.7

 Table 5. RPE scores of the soldiers during exercise fatigue evaluation

CF: compressed food; CFMO: compressed food with maltooligosaccharides; CFSO: compressed food with soy oligopeptides. Data are shown as mean±SE.

Body fatigue during exercise and endurance training is closely related to the increased blood lactate.¹⁵ The present study showed lower blood lactate levels with the CFMO diet compared with the other diets, which could be because of the added MO in the CFMO diet. In addition, the blood lactate clearance rate with the CFMO diet was the highest, followed by the CFSO diet, indicating the role of oligopeptides and oligosaccharides to speed up the removal of blood lactate produced during exercise. Faster lactate clearance helps to relieve fatigue quickly and promote physical recovery.

Increased blood hematocrit is correlated with RBC or the release of cells stored in blood sinus. Increased RBC can maintain the supply of oxygen and enhance exercise performance.^{16,17} Therefore, the increased RBC and hematocrit levels immediately after exercise with the CFSO diet may be helpful for oxygen supply during exercise. With the CFSO diet, hematocrit levels were higher than with the other diets, possibly because of extra protein supplements in this diet. Under normal circumstances, the body does not store protein. Extra proteins are metabolized by ammonia decomposition and the nitrogen is excreted in the urine. This process requires a lot of water that leads either to body over-hydration or water deficit state, which increases hematocrit.

Urine specific gravity is often used to assess hydration status. The measurement of urine specific gravity and pH may be useful for the estimation of events in the body after submaximal exercise loading.18 The present study showed that the concentration of body fluids with the CFMO diet was significantly lower than with the other diets. Therefore, the CFMO diet could maintain urine pH and avoid acidification of body fluids. A decrease in urine pH with the CFSO diet was observed immediately after exercise. These findings suggest that acidic substances (such as sulfate or phosphate) produced during protein metabolism led to lower urine pH with the CFSO diet due to its high protein content. Decreased rate of gastric emptying because of high protein content may reduce the rate of influx of water into the circulation, resulting in the reduction of urine production.¹⁹ Decreased urine production leads to an increase in urine specific gravity due to smaller dilution. A protein-enriched diet could increased water absorption.²⁰ From these results, it can be inferred that there may be a tendency for increased fluid retention with the CFSO diet. Fluid retention is the main index for assessing rehydration effectiveness. In addition, the CFSO diet could be more effective for rehydration compared with the CFMO diet.

Diets and daily training may affect liver function.^{21,22}

Four common parameters to test liver function (serum TP, ALB, ALT, and AST) were analyzed in the present study. Results showed that the consumption of CF did not result in decreased liver function.

Strenuous physical exertion in hot environments can result in acute kidney injury.²³ Creatinine is the final product of creatine metabolism in muscle, and is excreted primarily by glomerular filtration. Serum creatinine levels are used to assess renal function.²⁴ Creatine supplementation minimally affects creatinine and renal function in young healthy adults and does not enhance exercise performance.^{25,26} In the present study, no significant difference in serum creatinine was observed among all diets. During continuous exercise, the energy balance in muscle is destroyed, protein and amino acid catabolism increases, urea production increases, and the generated urea cannot be excreted. All of these factors can result in elevated blood urea levels.²⁷ Exercise also results in increased sweating, decreased urine production, and increased blood urea. In addition, protein intake can also affect blood urea levels.^{28,29} In the present study, the BUN of the participants with the CFSO diet was significantly higher than with the other diets. The lowest BUN was observed at each time point with the CF diet. The BUN with the CFSO and CF diets may be highly associated with the protein content of these diets. Purine metabolism is one of the most important ways of excreting nitrogenous waste. UA is the end product of purine metabolism in the body.³⁰ All UA is filtered by the glomeruli, and approximately 98-100% of UA is reabsorbed in the proximal tubule. Therefore, under normal circumstances, the clearance rate of UA is low. However, UA levels may increase after daily training. It has been reported that urine UA is increased after exercise.³¹ Profuse sweating during exercise could result in decreased urinary UA excretion and lead to increased serum UA after exercise.³² In the present study, the UA with the CFSO diet was significantly higher compared with the other diets. High blood UA with the CFSO diet could be associated with high protein content, similar to BUN.

CK is mainly found in skeletal muscles, cardiac muscles, and brain tissues. Changes of CK can be used as an indicator for evaluating the loading, injury, adaptation, and recovery of skeletal muscle.^{33,34} Myoglobin is a small protein that is mainly found in skeletal muscles. Its main function is to store a small amount of oxygen for use by the body during hypoxia.³⁵⁻³⁷ TnI is a contractile protein found in the cardiac muscle. During cardiac muscle damage, TnI significantly increases and TnI is used as a specific and sensitive serum marker for diagnosing myocardial damage.^{38,39} The results of the present study suggested that the consumption of CFMO and CFSO for 4 consecutive days had no obvious effects on preventing skeletal or cardiac muscle damage induced by training. These findings suggest that protein and carbohydrate supplementation could not alter intramuscular anabolic signaling response after resistance exercise.⁴⁰ Another study showed that short-term supplementation of amino acids produced by protein hydrolysis before and during a 100km ultra-marathon had no effect on skeletal muscle damage and muscle soreness.⁴¹ Other studies showed that protein supplementation had no effect on muscle damage,⁴² and that carbohydrate supplementation during recovery from soreness-inducing exercise did not promote the muscle responses.⁴³

The RPE was developed to provide a subjective estimation of exercise intensity.44 The RPE is a recognized marker of intensity and of homeostatic disturbance during exercise and is typically monitored during exercise tests to complement other measures of intensity.⁴⁵ The present study showed no significant difference in RPE scores among the four diets during exercise. Another study showed no change in RPE during tennis performance between high-carbohydrate and placebo diet groups.⁴⁶ Kerasioti et al reported that the mean RPE was not significantly different between subjects consuming a special cake consisting of carbohydrates and whey protein and subjects consuming an isocaloric carbohydrate cake during exercise.⁴⁷ Duckworth et al also demonstrated that there were no significant differences in RPE among subjects consuming galactose, glucose, or placebo for overall RPE or RPE at specific time points assessed during moderate intensity exercise.48

This study has some limitations. We only included 12 participants; future studies with a larger number of participants would add more weight to these results. Additionally, we studied consumption of CF over a short time period, but it may take a longer time to show significant effects on some parameters such as liver and renal functions or muscle injury. The small sample size can be a limitation of this study, but a similar study that evaluated the efficacy of carbohydrate and protein diet on endurance had been conducted with a small sample size and results could be used as a basis for future studies.⁴

In conclusion, the CFMO diet may lead to better recovery of HR, improve and maintain blood glucose levels, and increase the removal of blood lactate produced during fatigue without acidification of body fluids. The CFSO diet may have advantages in accelerating removal of blood lactate during recovery, maintaining oxygen supply, and augmenting fluid retention. Based on the present study, it can be inferred that MO and SO could maintain and improve exercise performance in military endurance trainings. At present, the design and manufacture of CF is not only focused to supply energy and nutrition, but also to enhance functions, especially improvement in exercise ability, which is the most important factor for soldiers to carry out military tasks efficiently. Therefore, the supplement of military food with functional objectives to enhance exercise performance should be further explored.

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AUTHOR DISCLOSURES

All the authors declare that they have no conflict of interest.

REFERENCES

- 1. Arya SS. Foods for the armed forces: global scenario. Mysore, India: Defence Food Research Laboratory; 1998.
- Azanza MP. Canned rice products as Philippine military food ration. Int J Food Sci Nutr. 2003;54:235-40. doi: 10. 1080/09637480120092008.
- Hirsch ES, Matthew Kramer F, Meiselman HL. Effects of food attributes and feeding environment on acceptance, consumption and body weight: lessons learned in a twentyyear program of military ration research US Army Research (Part 2). Appetite. 2005;44:33-45. doi: 10.1016/j.appet.2004. 04.010.
- Ivy JL, Res PT, Sprague RC, Widzer MO. Effect of a carbohydrate-protein supplement on endurance performance during exercise of varying intensity. Int J Sport Nutr Exerc Metab. 2003;13:382-95.
- Ohta A. Prevention of osteoporosis by foods and dietary supplements. The effect of fructooligosaccharides (FOS) on the calcium absorption and bone. Clin Calcium. 2006;16: 1639-45. doi: CliCa061016771684. (In Japanese)
- Yoshii H, Tachi N, Ohba R, Sakamura O, Takeyama H, Itani T. Antihypertensive effect of ACE inhibitory oligopeptides from chicken egg yolks. Comp Biochem Physiol C Toxicol Pharmacol. 2001;128:27-33.
- Cai MY, Gu RZ, Li CY, Ma Y, Dong Z, Liu WY, Jin ZT, Lu J, Yi WX. Pilot-scale production of soybean oligopeptides and antioxidant and antihypertensive effects in vitro and in vivo. J Food Sci Technol. 2014;51:1866-74. doi: 10.1007/s13197-012-0701-4.
- Shiraga T. Effect of a soy-derived oligopeptide diet on plasma levels of endogenous morphine-like analgesic peptides in rats. Soy Protein Research, Japan. 2007;10:59-62. (In Japanese)
- Yaspelkis BB, 3rd, Patterson JG, Anderla PA, Ding Z, Ivy JL. Carbohydrate supplementation spares muscle glycogen during variable-intensity exercise. J Appl Physiol. 1993;75: 1477-85.
- Baty JJ, Hwang H, Ding Z, Bernard JR, Wang B, Kwon B, Ivy JL. The effect of a carbohydrate and protein supplement on resistance exercise performance, hormonal response, and muscle damage. J Strength Cond Res. 2007;21:321-9. doi: 10.1519/R-21706.1.
- 11. Borg G. Borg's perceived exertion and pain scales. Champaign, IL: Human Kinetics. 1998.
- Borresen J, Lambert MI. Changes in heart rate recovery in response to acute changes in training load. Eur J Appl Physiol. 2007;101:503-11. doi: 10.1007/s00421-007-0516-6.
- Khanna GL, Manna I. Supplementary effect of carbohydrate-electrolyte drink on sports performance, lactate removal & cardiovascular response of athletes. Indian J Med Res. 2005;121:665-9.
- Foskett A, Williams C, Boobis L, Tsintzas K. Carbohydrate availability and muscle energy metabolism during intermittent running. Med Sci Sports Exerc. 2008;40:96-103. doi: 10.1249/mss.0b013e3181586 b2c.
- 15. Gorostiaga EM, Navarro-Amezqueta I, Gonzalez-Izal M, Malanda A, Granados C, Ibanez J, Setuain I, Izquierdo M. Blood lactate and sEMG at different knee angles during

fatiguing leg press exercise. Eur J Appl Physiol. 2012;112: 1349-58. doi: 10.1007/s00421-011-2090-1.

- 16. Gonzalez-Alonso J. ATP as a mediator of erythrocytedependent regulation of skeletal muscle blood flow and oxygen delivery in humans. J Physiol. 2012;590:5001-13. doi: 10.1113/jphysiol.2012.235002.
- Page TC, Light WR, Hellums JD. Prediction of microcirculatory oxygen transport by erythrocyte/ hemoglobin solution mixtures. Microvasc Res. 1998;56:113-26. doi: 10.1006/mvre.1998.2088.
- Moriguchi T, Shimomitsu T, Odagiri Y, Fukuda J, Hamano K, Kawai T, Tomoda A. Marked increase in urinary bicarbonate and pH caused by heavy muscular exercise with dynamic knee extension. Tohoku J Exp Med. 2002;198:31-9.
- Evans GH, Shirreffs SM, Maughan RJ. The effects of repeated ingestion of high and low glucose-electrolyte solutions on gastric emptying and blood 2H2O concentration after an overnight fast. Br J Nutr. 2011;106:1732-9. doi: 10. 1017/S0007114511002169.
- Lauridsen TG, Vase H, Starklint J, Bech JN, Pedersen EB. Protein-enriched diet increases water absorption via the aquaporin-2 water channels in healthy humans. Nephrol Dial Transplant. 2010;25:2502-10. doi: 10.1093/ndt/gfq111.
- 21. Kinoshita S, Tsuji E. The effect of different intensities of exercise on plasma and liver alpha-tocopherol levels and hepatic thiobarbituric acid reactive substance levels of rats. Jpn J Phys Fit Sport. 2001;50: 445-51.
- 22. Wijck K, Lenaerts K, van Loon LJ, Senden JM, Buurman WA, Dejong CH. Increased small intestinal permeability and liver damage after exhaustive physical exercise in healthy individuals. Gastroenterology. 2011;140:S-647.
- 23. Junglee NA, Di Felice U, Dolci A, Fortes MB, Jibani MM, Lemmey AB, Walsh NP, Macdonald JH. Exercising in a hot environment with muscle damage: effects on acute kidney injury biomarkers and kidney function. Am J Physiol Renal Physiol. 2013;305:F813-20. doi: 10.1152/ajprenal.00091.20 13.
- Stark J. Interpretation of BUN and serum creatinine. An interactive exercise. Crit Care Nurs Clin North Am. 1998;10: 491-6.
- Mendes RR, Tirapegui J. Physical exercise, creatine and nutrition: a review. Revista Brasileira de Ciencias Farmaceuticas. 1999;35:195-209.
- Pline KA, Smith CL. The effect of creatine intake on renal function. Ann Pharmacother. 2005;39:1093-6. doi: 10.1345/ aph.1E628.
- Mohammed A, Mohammed G, Akerejola O. Haematological and serum biochemical changes in Bunaji work bulls after farmland ridging exercise in Kaduna State, Nigeria. J Anim Vet Adv. 2007;6:576-9.
- Colombini A, Machado M, Lombardi G, Lanteri P, Banfi G. Modifications of biochemical parameters related to protein metabolism and renal function in male soccer players after a match. J Sports Med Phys Fitness. 2014;54:658-64.
- Wickens CL, Moore J, Shelle J. Nitrogen balance and blood urea nitrogen as response criteria to estimate protein requirement of the exercising horse. J Anim Sci. 2003;81:48.
- 30. Guerreiro S, Ponceau A, Toulorge D, Martin E, Alvarez-Fischer D, Hirsch EC, Michel PP. Protection of midbrain dopaminergic neurons by the end-product of purine metabolism uric acid: potentiation by low-level depolarization. J Neurochem. 2009;109:1118-28. doi: 10. 1111/j.1471-4159.2009.06040.x.
- Xing X, Shi X, Zhang M, Jin W, Ye J. CE determination of creatinine and uric acid in saliva and urine during exercise. Chromatographia. 2008;67:985-8.

- Huang LL, Huang CT, Chen ML, Mao IF. Effects of profuse sweating induced by exercise on urinary uric acid excretion in a hot environment. Chin J Physiol. 2010;53:254-61.
- Brancaccio P, Maffulli N, Limongelli FM. Creatine kinase monitoring in sport medicine. Br Med Bull. 2007;81-82:209-30. doi: 10.1093/bmb/ldm014.
- 34. Havas E, Komulainen J, Vihko V. Exercise-induced increase in serum creatine kinase is modified by subsequent bed rest. Int J Sports Med. 1997;18:578-82. doi: 10.1055/s-2007-972 684.
- 35. Papadopoulos S, Endeward V, Revesz-Walker B, Jurgens KD, Gros G. Radial and longitudinal diffusion of myoglobin in single living heart and skeletal muscle cells. Proc Natl Acad Sci U S A. 2001;98:5904-9. doi: 10.1073/pnas.10110 9798.
- Richardson RS. Oxygen transport: air to muscle cell. Med Sci Sports Exerc. 1998;30:53-9.
- 37. Spires J, Lai N, Zhou H, Saidel GM. Hemoglobin and myoglobin contributions to skeletal muscle oxygenation in response to exercise. Adv Exp Med Biol. 2011;701:347-52. doi: 10.1007/978-1-4419-7756-4_47.
- Akyuz M. Changes in serum cardiac troponin T levels in professional football players before and after the game. Afr J Pharm Pharmaco. 2011;5:1365-8.
- 39. Shave R, Baggish A, George K, Wood M, Scharhag J, Whyte G, Gaze D, Thompson PD. Exercise-induced cardiac troponin elevation: evidence, mechanisms, and implications. J Am Coll Cardiol. 2010;56:169-76. doi: 10.1016/j.jacc. 2010.03.037.
- 40. Gonzalez AM, Hoffman JR, Jajtner AR, Townsend JR, Boone CH, Beyer KS et al. Protein supplementation does not alter intramuscular anabolic signaling or endocrine response after resistance exercise in trained men. Nutr Res. 2015;35:990-1000. doi: 10.1016/j.nutres.2015.09.006.
- 41. Knechtle B, Knechtle P, Mrazek C, Senn O, Rosemann T, Imoberdorf R, Ballmer P. No effect of short-term amino acid supplementation on variables related to skeletal muscle damage in 100 km ultra-runners - a randomized controlled trial. J Int Soc Sports Nutr. 2011;8:6. doi: 10.1186/1550-2783-8-6.
- Lee J. Effect of protein supplementation on exercise-induced muscle damage. Kor J Sport Sci. 2010;21:1298-314.
- 43. Miles MP, Pearson SD, Andring JM, Kidd JR, Volpe SL. Effect of carbohydrate intake during recovery from eccentric exercise on interleukin-6 and muscle-damage markers. Int J Sport Nutr Exerc Metab. 2007;17:507-20.
- 44. Kilpatrick MW, Robertson RJ, Powers JM, Mears JL, Ferrer NF. Comparisons of RPE before, during, and after selfregulated aerobic exercise. Med Sci Sports Exerc. 2009;41: 682-7. doi: 10.1249/MSS. 0b013e31818a0f09.
- Eston R. Use of ratings of perceived exertion in sports. Int J Sports Physiol Perform. 2012;7:175-82.
- 46. Gomes R, Viveiros L, Coutts A, Ugrinowistch C, Aoki M. Effect of carbohydrate supplementation on tennis match play performance. J Sci Med Sport. 2010;12:e122.
- 47. Kerasioti E, Kiskini A, Veskoukis A, Jamurtas A, Tsitsimpikou C, Tsatsakis AM, Koutedakis Y, Stagos D, Kouretas D, Karathanos V. Effect of a special carbohydrateprotein cake on oxidative stress markers after exhaustive cycling in humans. Food Chem Toxicol. 2012;50:2805-10. doi: 10.1016/j.fct.2012.04.015.
- Duckworth LC, Backhouse SH, Stevenson EJ. The effect of galactose ingestion on affect and perceived exertion in recreationally active females. Appetite. 2013;71:252-8. doi: 10.1016/j.appet.2013.08.009.