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SHORT FAT-ADAPTATION DIET IMPACT ON A CONSECUTIVE DAY OF INTERVAL EXERCISE PERFORMANCE

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ABSTRACT

Background. During competition season, races and games can be scheduled multiple times a week or even within 24 hours. This may interfere with macronutrient periodization, carbohydrate loading regimen, hydration status and nutrition. Most of the studies investigating the influence of diet on performance do not take into consideration that an athlete may need to perform closely spaced, repeated events. The study tested whether the fat-adaptation diet would improve performance on the consecutive day of interval exercise.

Methods. Nine healthy, male amateur athletes were randomly assigned to two diets in a single-blinded, crossover study. The fat-adaptation diet consisted of the first five days high-fat diet (2.62 g/kg/d carbohydrates, 2.23 g/kg/d fat). The day six and seven of the fat-adaptation diet consisted of 5.42 g/kg/d carbohydrates. The balanced carbohydrate diet consisted of a seven-day protocol involving consumption of 5.33 g/kg/d. On day seven of each diet protocol, subjects performed an interval treadmill test dependent on exhaustion. Blood glucose and lactate were measured before and immediately after exercise. The identical treadmill test was performed once again after 24 hours on the day eight of each diet.

Results. There was a significant decrease in the total distance to exhaustion after the fat-adaptation diet (11.2 \pm 0.6 km vs 10.9 \pm 0.8 km), p < 0.05 with lactate being lower after exercise on the second day (6.2 \pm 0.8 mM) compared to the first day (7.4 \pm 0.9 mM). Glucose was elevated after exercise except on the second test day on the fat-adaptation diet (5.3 \pm 0.3 mmol/L).

Conclusions. Athletes perform better on the balanced carbohydrate diet than short fat-adaptation diet on the consecutive day of interval test.

Keywords: human nutrition, exercise performance, fat adaptation, carbohydrate, macronutrients

INTRODUCTION

The position statement from the American College of Medicine, Academy of Nutrition and Dietetics, and Dietitians of Canada for athletes is to consume 6–10 g of carbohydrate per kilogram of body weight daily (Thomas et al., 2015). High amounts of carbohydrates as the main energy source for the athletes has been shown to have many benefits. Fatigue at the end of an event may be due to glycogen depletion (Bangsbo et

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al., 2007). The decision on how much carbohydrates an athlete should consume depends upon the type of exercise in which the athlete participates and time of the season. Athletes change the intensity of their training to ensure appropriate glycogen storage (Burke et al., 2007). It may be very difficult to apply all nutrition recommendations, especially when we take into consideration the glycogen depletion exercise since most athletes taper their exercise routinely before the big competition. During competition season, very often races and games are scheduled multiple times a week or even within 24 hours and involve a lot of travelling. This may interfere with carbohydrate loading regimen, hydration status and nutrition. Most of the studies investigating the influence of diet on performance do not take into consideration that an athlete may need to perform closely spaced, repeated events what could be the reason why some supplemental regimen do not work in laboratory setting (Główka and Woźniewicz, 2019).

Optimizing utilization of glycogen stores during competition by using a low carbohydrate diet is one of the researched methods for optimum performance. However, this method is contrary to what has been used among athletes and the theory of carbohydrate loading (Rapoport, 2010). A low carbohydrate diet is associated with increased muscle enzymes activity responsible for beta-oxidation/transport of fatty acids and an increase in gluconeogenesis. To date, there is little evidence that training in a glycogen depleted state improves athletic performance (Burke et al., 2017; Chang et al., 2017). Most of the benefits are only visible on the cellular levels. It is likely that an athlete with low glycogen stores will not perform at his or her best capabilities (Atkinson et al., 2011; Burke et al., 2017; Impey et al., 2018).

Another type of diet manipulation for glycogen depletion, termed fat adaption, uses a short- term low-carbohydrate diet (training phase) followed by 24 hours of glycogen restoration through the intake of a high carbohydrate diet one day before the competition. Therefore, it is possible that cell adaptations to glycogen depletion caused by the diet will remain even after short term carbohydrate loading. More studies are needed in order to see if fat adaptation improves performance.

There have been no studies on fat adaptation diets among team athletes that consider the long-term effect of a low carbohydrate diet and training on a fasted state, or a short-term low carbohydrate diet with glycogen repletion. In one of the study, concerning cross-fit athletes, the ketogenic diet seemed to improve fat oxidation during moderate intensity exercise (Durkalec-Michalski et al., 2019). However, the unknown is if these adaptations are enough to overcome the glycogen depletion and have a positive impact on performance. The purpose of this study is to test the hypothesis that fat adaptation diet will improve repetitive performance after a treadmill exercise test.

METHODS

Study participants

A total of nine male amateur athletes (weight: 72.5 ± 2.9 kg, age: 29.5 ± 3 y, body fat: 8.3 $\pm 0.7\%$) finished the experiment approved by the local Health Sciences Institutional Review Board. All subjects were recruited from flyers posted around the University and emails to the local coaches. The subjects had to be participating in an amateur sport requiring intense physical activity and have at least four training units per week. After the procedures, risks, and benefits of the study were explained, each subject signed informed consent. American College of Sports Medicine screening guidelines were used to assure only low-risk subjects participated in the study (Green, 2010). Body fat percentage was measured by bioelectrical impedance method (Tanita Inc., MC-58). After the screening, subjects were familiarized with the protocol, which included performing the interval running a test on a treadmill. Subjects had to finish at least the 67th minute of test (9.84 km) to be included in the study. Eleven subjects were accepted into the study; two resigned due to difficulties with adhering to the diets.

Diet design

The study compared two iso-caloric diets: fat adaptation (fat-adapt) and balanced carbohydrate (BCHO). The fat-adapt consisted of the first 5 days being high in fat and low carbohydrate followed by two days of a balanced carbohydrate diet. The BCHO protocol consisted of all 7 days being balanced in carbohydrates. The diet composition acquired from dietary records is described in Table 1. Diet assignment and its analysis after the study were done by using software NutritionPro 5.0 (Axxya, Stafford, Texas). Diet design was



Fig. 1. Repetitive exercise study timeline. Day 1 was screening visit and familiarization with the treadmill protocol. On day 1, the subject started the assigned diet: fat adaptation (Fat adapt) or balanced carbohydrate (BCHO) and came back after a week for visit 2 (day 7) and 3 (day 8). After visit 3, the subject started a washout period for 7 days – returning to habitual diet. Diet 2 started following the washout. Set of two testing visits is repeated after diet 2. During the visit subjects had their capillary blood lactate and glucose measured. After the treadmill protocol subjects had 3 minutes to cool down at 4.8 km/h blood lactate and glucose were taken immediately after

based on the subject's weight and age. The dietitian considered subject's food preferences (e.g.: likes and dislikes, home cooking, or eating outside) to facilitate the dietary adherence. Diet design was based on Dietary Guidelines for Americans 2010 (McGuire, 2011). The last meal before the stress test was the same before each visit. Within 30 minutes after each study test, a subject had to drink 350 ml of sport drink which provided 20 g of carbohydrates. Subjects were asked to abstain from alcohol. The subjects were advised to not participate in any tournaments during the study length and proceed with similar training intensity as a week before visit 1. The diet order was assigned randomly (Fig. 1). At the end of first visit, each subject met with a Registered Dietitian (RD) to discuss the first diet. The lab staff who performed the testing was blinded to the diet. Subjects started the diet after the day of screening and kept it for 7 days. On day 7, subjects came fasting. Within 24 hours after the visit on day 7,

subjects came back to repeat the testing protocol. All subjects were assigned to have a high carbohydrate diet on day 7 and to come back on the day 8th fasted. After the first diet and two days of testing, the subjects started a one-week habitual diet washout period. Next, the subjects started the second diet. After receiving the prescribed 7 day menu, subjects stayed in touch with the RD. To check the dietary compliance, the subjects did 7 day dietary records during each intervention. All the food diaries were checked and consulted with the RD during the visit.

Lactate and glucose measurements

Capillary blood for lactate (Accutrend lactate meter, Roche Mannheim Germany) and glucose (AccuCheck Aviva glucometer, Roche Mannheim Germany) was collected from the ear immediately before the test and after the 3 minutes of cool down at 3 mph following the test.

Treadmill protocol

The test protocol is adapted to treadmill version of Yo--Yo endurance intermittent test preceded by the warmup (Bangsbo and Lindquist, 1992). The warm-up consisted of the continuous sequential 6 minute runs at 6.5, 8, 9.5 km/h with a 2-minute rest between each run. After that, the subjects ran for 3 minutes at 14.5 km/h followed by a 3-minute rest. At the 30th minute, the intermittent portion of the exercise started, which consisted of 5 minute runs at two alternating speeds (15 s at each speed, repeated): 8/10.5 km/h, with a 2-minute rest, 8/11, 8/12, 8/13, 9.5/14.5, 11/17, 14.5/17 and a 5-minute rest between each. After a final 5-minute rest, subjects ran at 17 km/h until voluntary exhaustion. After the test, each subject walked at 3 mph for 3 minutes. The subject's heart rate and Rating of Perceived Exertion (RPE) were taken before the start and after completion of each interval (Dunbar et al., 1992) as well after a 3 minute after the test. Subject's performance on the treadmill was defined as time to exhaustion as well as the distance completed.

Statistics

The data was normalized before statistical analysis with repeated measures two-way ANOVA and three-way ANOVA (Sigmastat 3.5 software, Systat Software, San Jose, CA). Values are expressed as mean ±standard error of the mean. The primary outcome measure was performance measured as distance completed in km. Independent variables for repeated measures two-way ANOVA were the diet manipulation (fat-adapt vs BCHO), tests within 24 hours (test 1 vs test 2; repeated measures two-way ANOVA). The blood lactate and blood glucose were compared by repeated measure three-way ANOVA (before vs post, test 1 vs 2, diet BCHO vs fat-adapt). Blood lactate and glucose values were also analyzed separately before and after the exercise test (repeated measures two-way ANOVA). The significance was set to P < 0.05. Heart rate during the exercise and after exercise was analyzed by two-way ANOVA. Post hoc Holm-Sidak test was performed to analyze the interactions between the data within the subjects.

RESULTS

Daily intake of fatty acids, carbohydrates, and protein of each subject participating in the study were analyzed (Table 1). Carbohydrate intake on the BCHO diet versus the last 2 days of fat adaptation diet was not different statistically. The diets were iso-caloric with the percent of energy from carbohydrates being 65% and 30% with the fat percentage being 21% and 58% on the BCHO and fat-adapt diets, respectively. During the two days of glycogen repletion, the percentages of carbohydrates and fat were 64% and 24%, respectively.

Table 1. Daily nutrient intake during high carbohydrate and fat adaptation periods

Nutrient intake per day	Fat adaptation		Balanced carbohydrate
	5 days	2 days	7 days
Energy, kcal	$2510 \pm \! 96$	$2467 \pm \! 183$	$2370\pm\!\!117$
Protein, g	90 ± 6	$82\pm\!\!5$	90 ± 4
Carbohydrate, g	$190 \pm \! 10^{\rm a}$	$393\pm31^{\text{b}}$	$387 \pm \! 18^{\text{b}}$
Fat, g	$162 \pm 8^{\rm a}$	66 ± 8^{b}	$56\pm5^{\mathrm{b}}$

^{a, b}Values with common letters are not different, P < 0.05. Values are the mean ±the SEM of 9 subjects.

The participant's time to volitional exhaustion was measured during each visit. The distance covered was compared between each other and the total of visit 1 and 2 after the BCHO and fat-adapt (Fig. 2). The average total distance to exhaustion after the BCHO was significantly longer (11.2 ± 0.6 km) than the fat-adapt (10.9 ± 0.8 km) P < 0.05, F = 5.9. There was no difference between the two diets on the first day (fat adapt: 11.1 ± 0.7 km vs BCHO: 11.3 ± 0.7 km) P = 0.53, F = 0.43. The difference between the diets on the second day taken after 24 hours was not statistically significant (P = 0.057, fat adapt: 10.7 ± 0.6 km, BCHO: 11.1 ± 0.7 km).

Rating of Perceived Exertion – RPE was related to the testing protocol speeds. Heart rate was related to testing protocol speeds during every test.

Heart rate was measured 3 minutes after a 3 mph cool down period after the treadmill test. The heart rate was lower after test 2 of each diet (P < 0.05, F = 5.0; Table 2). The heart rate 3 minutes after test 1 of fat-adapt was lower after test 2 on the same diet (Table 2).

Blood lactate was significantly elevated after each exercise test. There was no difference detected between



Fig. 2. Distance to exhaustion. Each line pattern represents one subject and their performance to exhaustion in meters. Letters **a** and **b** represent a significant difference in performance between the diets with the tests combined, P = 0.041. Subjects run further on the balanced carbohydrate (BCHO) diet. Means are marked on the graph for each visit being 11 070 ±652 m, 10 664 ±650 m for fat-adapt, and 11 300 ±562 m, 11 118 ±663 m for BCHO visits 1 and 2 respectively

blood lactate and different testing visits. However, lactate was lower on the first test visit before the treadmill test after the fat-adapt diet compared to the second

Table 2. Capillary glucose and lactate, heart rate measured pre and post exercise

Parameter	Fat adaptation		Balanced carbohydrate	
	day 1	day 2	day 1	day 2
Glucose _{pre exercise} mmol/L	4.8 ± 1	$4.9\pm\!0.6$	4.7 ±1	4.7 ± 0.7
Glucose _{post exercise} mmol/L	5.8 ± 0.3	5.3 ±0.3°	5.8 ± 0.4	5.6 ± 0.3
Lactate _{pre exercise} mM	1.4 ±0.2°	2.1 ±0.2	2.0 ± 0.3	2.1 ±0.1
Lactate _{post exercise} Mm	$7.4 \pm \! 0.8$	6.2 ±0.8°	7.2 ± 0.7	7.0 ± 0.9
HR maxª, bpm	181 ± 5	184 ±4	178 ± 3	179 ± 4
HR rest ^b , bpm	124 ±3°	$119\pm\!\!3$	122 ± 3	$118 \pm \! 3$

^aMaximal heart rate measured at the end of exercise.

^bHeart rate measured after 3 minute rest after exercise.

[°]Represents a significant difference p < 0.05.

visit after both diets and visit 1 after BCHO (Table 2). After the fat-adapt, post-exercise lactate was lower on the second visit (6.2 ± 0.8 mM) than on the first visit (7.4 ± 0.9 mM). Blood glucose was elevated after exercise on all tests except test 2 of fat-adapt diet (Table 2).

DISCUSSION

The subjects ran longer on average over the two days on the balanced carbohydrate diet than on fat adaptation diet. The study does not support the hypothesis that adaptation to a high-fat diet will improve performance.

It is important to note that long term high fat diet may be harmful for athletes. Athletes training on a high-fat diet and using glycogen depletion techniques will train at lower capacities, therefore they may not prepare for the season (Mujika and Burke, 2011). Side effects from the high fat diet may be more troublesome than the ones from high carbohydrates: jittery, weakness, gastrointestinal problems may cause athlete to be less motivated during training sessions (Mujika and Burke, 2011). With this in line, improperly balanced diets are one of potential risks of health problems in athletes (Łagowska and Jeszka, 2011). To minimize the risk, our fat adaptation diet had increased carbohydrate intake to 21% of energy per day and compared it to macronutrient balanced diet. This might cause the cellular adaptations did not occur. However, the subjects reported difficulties with adhering to high fat diet.

This study is one of a few studies analyzing the influence of diet on an athlete's performance on consecutive days and using a high-intensity interval exercise protocol (Akermark et al., 1996; Cochran et al., 2015; Larsen et al., 2019; Rehrer et al., 2010). Even though, there are noticeable cellular adaptations after the low carbohydrate diet (Larsen et al., 2019), it is the appropriate amount of carbohydrate intake what improves the performance (Akermark et al., 1996; Vogt et al., 2005). In our study, the performance was trended to be better on the second day of balanced carbohydrate diet, what could be caused by good carbohydrate repletion. Team athletes, or cyclists have tournaments on the consecutive days and recovery is crucial for winning or losing the games. The use of the second day of testing within 24 hours may be useful to analyze the influence of different variables on the recovery of an athlete in the laboratory. In this study, the use of the second day of testing produced more interesting results. The distance to exhaustion was lower on the second day, regardless of the diet, and therefore it strengthened the power of the analysis of the total time to exhaustion. Blood lactate and glucose were significantly different based on diet, but only on the second day of testing.

The consecutive day of testing brought another interesting result: heart rate at many points during the testing as well as 3 minutes after the test decreased consistently. These differences may be due to the subject's familiarity with the test or fluid balance and hydration. The subject's water intake was measured during the test, and there was no difference in fluid intake between the visits (data not shown), it is possible that the first testing visit could impact fluid needs for optimal recovery or alter fluid consumption during the 24-hour recovery when they had free access to water. However, as expected there was no difference in the maximal heart rate between the visits, and at the same time points of the test the workload was the same, but the heart rates were still lower on the second day. Heart rate at the time-points during testing was also lower after the BCHO diet. The correlation of heart rate and the high carbohydrate diet have been investigated in the study involving twelve healthy males performing high and low-frequency tests on high carbohydrate diet and low carbohydrate diet (Lima-Silva et al., 2010). After the high carbohydrate diet, the heart rate during intense exercise was lower than after low carbohydrate diet. The blood catecholamines were also measured in the study and there was no difference in concentration between the two diets. The authors of the study speculated that the difference in heart rate may be due to autonomic control. The difference between heart rates and two diets may support the beneficial effects of high carbohydrate diets on performance.

As expected, ratings of perceived exertion did show a correlation with the heart rate during the performance. Rating of Perceived Exertion – RPE taken at the end of each interval was at many time points significantly higher on the day two after the fat-adapt diet, which suggests that subjects were not completely recovered from the day one test. Lima-Silva et al. (2010) compared the RPE after two days of high fat and high carbohydrate diet. The results are similar to our results with the time to exhaustion and the rate of increase of RPE being lower on the high fat diet.

Lactate measured before exercise was lower on the first visit of the fat-adapt diet when compared with all other visits. Lactate measured after exercise on the second day of fat-adapt diet was also lower than lactate measured after other testing visits. These results may be associated with cellular adaptations. Blood glucose was elevated after exercise in all cases except on the second test on the fat-adapt diet. This difference may have been caused by either that the fatadapt diet decreased liver glycogen storage with the second day of testing being the most affected, or that there was more muscle glucose uptake due to a lack of muscle glycogen. In the other studies investigating psychological and physiological responses to exercise and high carbohydrate diet, those measurements were very variable. In the study by Lima-Silva et al. (2011), there was no significant influence of diets on glucose or lactate. It is possible that this difference in results was caused by the length of the diet and carbohydrate content and the type of exercise testing.

The results of our study do not agree with the fat adaptation theory. High-fat diet cause higher gene expression of fatty acid translocase and beta-hydroxyacyl-CoA (Cameron-Smith et al., 2003). However in the more recent study, there was no difference shown in the mRNA response of markers of mitochondrial biogenesis with carbohydrate restriction (Jensen et al., 2015). It may seem that glycogen availability is more important for physical performance than cell adaptations caused by glycogen depletion before the physical testing, especially after a day of exhaustive exercise. This study design, however, allowed only for a shortterm high fat diet and no biopsies. Therefore we cannot state if any kind of cellular adaptations occurred. In the other study concerning fat adaptation followed by carbohydrate restoration, there was no difference in 2 hour performance between the two diets (Burke et al., 2000). However, the fat oxidation increased and carbohydrate oxidation decreased in muscle tissue after the 2-hour trial in the fat adaptation diet. Similar results were obtained in other studies (Carey et al., 2001; Havemann, 2006). In the study by Carey et al. (2001), performance on fat adaptation diet improved however, it was not statistically significant. In the experiment done by Havemann et al. (2006), there was a tendency

for power output during a 100 km time trial to decrease after a fat adaptation diet. The only study showing a significant performance improvement after a shortterm fat adaptation diet with carbohydrate loading is by Lambert et al. (2001). In another study glycogenolysis was noticed to be lower after the fat adaptation diet (Stellingwerff, 2005). Other studies show that the short-term high-fat diet does influence cell signalling and protein translocation (Cameron-Smith et al., 2003; Carey et al., 2001; Lambert et al., 2001; Stellingwerff, 2005). Our study protocol considered balanced carbohydrate diet (~6 g/kg/d) for comparison to fat adaptation diet. This might be not enough of carbohydrate to replete the glycogen stores. Diet with very high carbohydrate (>8 g/kg/d) could even more increase the difference in performance between the two diets. Although diet design was based on micronutrient intake, and seven day dietary record was taken to assess the dietary adherence, especially micronutrient intake may be not reliable. However, our study was short term and could not cause any micronutrient deficiencies. Subjects had access to the dietitian who helped them in keeping the assigned diet throughout the entire study. However, there would be less room for error if the subjects ate their meals from a metabolic kitchen. It would be valuable to see the rate of recovery at day 3 or 4 of consecutive day testing if the fat adaptation diet would cause even lower performance. Many athletes, for example, cyclists, compete throughout multiple consecutive days.

In conclusion, the study has shown the benefit of a high carbohydrate diet over a fat adaptation diet with carbohydrate restoration on consecutive days. More studies are needed to explore those benefits. The difference in the performance may be more detectable with a larger sample size. There is also a need in performing studies which test physical exertion on the consecutive days. This type of design may mimic closer the exertion which athletes experience during the competition season than a single day of exhaustive exercise.

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