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SUMMARY

Problem

The diet of military personnel is predicated on the basis that carbohydrate-rich diets delay the onset of fatigue. However, data concerning the effectiveness of carbohydrate-rich diets in delaying fatigue associated with short-term exercise is equivocal.

Objective

The object of this study was to correlate skeletal muscle fatigue profiles with cellular metabolites in exercising individuals who had high-fat or high-carbohydrate diets.

Approach

These studies measured quadriceps skeletal muscle pH, inorganic phosphate and high-energy phosphates using magnetic resonance spectroscopy (MRS) during graded exercise on subjects who had consumed high-carbohydrate or high-fat diets on a randomized cross-over fashion. Simultaneously respiratory and gas exchange data was collected during exercise periods and correlated with changes in cellular metabolites.

Results

A diet rich in carbohydrates prolongs incremental exercise time without affecting peak oxygen consumption and the improved exercise performance is related to relative preservation of skeletal muscle phosphocreatine and inorganic phosphate.

Conclusion

Diet influences exercise performance at least in part through changes in skeletal muscle high-energy phosphates. Carbohydrates likely improves short-term incremental exercise because of its inherent ability to rapidly generate more energy per mole of oxygen consumed. Reduced charbohydrate availability due either to depletion of glycogen stores or increased fat intake can compromise short-term exercise capacity.

INTRODUCTION

Since the last century, efforts have been made to delay the onset of fatigue by the manipulation of dietary composition. The beneficial effects of a diet high in carbohydrate (CHO) on endurance exercise performance were recognized in the 1930s (Christensen & Hansen, 1939). The suggestion that increased CHO intake improves exercise capacity via glycogen stores was first made nearly 30 years ago (Bergstrom et al., 1967; Hermansen et al., 1967; Hultman & Bergstrom, 1967). Conversely, deleterious effects of dietary fat on endurance exercise performance (Christensen & Hansen, 1939) have been described and related to decreased muscle glycogen stores (Hultman, 1989) and glycolytic flux (Jansson & Kaijser, 1982). Thus, for prolonged exercise, it is generally accepted that performance is largely related to the availability of carbohydrate substrate in the form of muscle glycogen.

Peripheral fatigue during intense exercise of short duration, however, may be due to other factors since muscle glycogen is relatively well preserved at exhaustion (Hermansen & Vaage, 1977). Putative mechanisms responsible for fatigue include depletion of high energy phosphates and accumulation of "toxic" metabolites such as Adenosine (ADP), hydrogen ion (H^+) and inorganic phosphate (P_i) (MacLaren et al., 1989).

Data concerning diet and short-term exercise capacity are relatively scanty and somewhat conflicting. Five groups of investigators found improved exercise time when intense exercise followed a CHO- vs. fat-rich diet (Greenhaff et al., 1987; Maughan & Poole, 1981; McLellan & Gass, 1989; White et al., 1988; Yoshida, 1986). Conversely, Quirion et al. found no change in incremental exercise time or peak workload following 48 hr of a high-CHO or high-fat diet (Quirion et al., 1988). These studies, however, employed indirect measurements of gas exchange to assess the influence of diet on short-term exercise performance. We recently developed a protocol which allows simultaneous noninvasive measurements of ventilation, gas exchange, and skeletal muscle metabolism by ^{31}P-magnetic resonance spectroscopy (MRS) during incremental quadriceps exercise. Since MRS can continuously measure skeletal muscle pH, P_i, and high-energy phosphates, it was used in the current study to better understand the intracellular

biochemical mechanisms linking dietary modifications to subsequent incremental exercise performance.

METHODS

Subjects. The study was approved by the Institutional Review Boards of the Massachusetts General Hospital (MGH) and the Massachusetts Institute of Technology (MIT). Inclusion criteria included a negative past medical history, normal physical examination, resting EKG and pulmonary function test. Five male and five female MGH employees and students (age=33 ± 3 years, height=169 ± 3.0 cm, and weight=66.6 ± 2.8 kg) were recruited for the study through bulletin board advertising. One female subject was unable to complete the study due to viral illness. Of the remaining 9 subjects, three jogged regularly and one was a competitive speed skater. Four of the remaining subjects walked 30 to 90 min per day. The last subject was sedentary. Subjects maintained their regular activity schedules throughout the study except on the two exercise study days.

Study Design. Maximal incremental quadriceps exercise was performed on each of two separate days, once following 5 days of high-carbohydrate (HCHO) diet and again after 5 days of a high-fat (HFAT) diet in a randomized, cross-over fashion. The controlled diets were separated by 2.5 days of ad libitum intake of a mixed diet.

Dietary Manipulation. The HCHO diet consisted of 75% CHO, 10% fat, and 15% protein. The HFAT dietary prescription included 70% fat, 15% CHO, and 15% protein. Eucaloric requirements were estimated for the two diets using a metabolic ward-prediction equation based on body weight and sex (unpublished data), adjusted for activity and rounded to the nearest 200 kcal.

On each morning of the dietary manipulation phases, subjects reported to the MGH General Clinical Research Center (GCRC). They were weighed in the fasting state after urination, while dressed in hospital gowns of known weight. The energy content of the diet was then adjusted as needed in 200 kcal units to maintain a body weight within 1.0% of that recorded

at the outset of the study. All foods were weighed and prepared for daily take-out. Subjects were encouraged to consume all of their food but instructed to return unconsumed portions to the GCRC the following morning. The weights of returned items were recorded. Daily energy, protein, fat, and CHO intake were calculated from actual weights of food consumed. Consumption of noncaloric sweeteners and habitual amounts of black coffee and tea were allowed.

On each exercise study day, subjects arrived at the MIT Francis Bitter National Magnet Laboratory between 0900 and 1700 hr having refrained from caffeine consumption and exercise. Subjects consumed a 600 kcal liquid meal of the same composition as the preceding diet, 3 hr before exercise. Subjects who exercised after 1400 hr on each day were also provided with their standard HFAT or HCHO breakfast at 0800.

Exercise procedure. Graded quadriceps exercise was performed in the supine position inside a 120 cm diameter bore MRS magnet (IBM/MIT, Cambridge, MA) and consisted of 90° bilateral leg extension against a padded bar resting over the distal legs at 0.5 Hz paced by a metronome (Fig. 1). After 1 min of exercise without imposed resistance, "workloads" were increased by 1.4 to 2.8 kg/min until exhaustion, defined as inability to maintain either the 0.5 Hz cadence paced by a metronome or a full-leg extension for three successive efforts. Exercise bouts were supervised by an investigator unaware of the subjects' dietary regimen.

Ventilatory and Gas Exchange Measurements. Respiratory and gas exchange data were collected continuously during a 5-min baseline period and throughout exercise. With noseclip and mouthpiece in place, subjects inspired room air through a 2-way J-valve (Collins Co., Braintree, MA). The expired gas volume ($\dot{V}E$), oxygen (O_2), and carbon dioxide (CO_2) fractions were measured breath-by-breath using a commercially available metabolic cart (SensorMedics Corporation 2900, Loma Linda, CA). The system was calibrated within factory specifications before each exercise run.

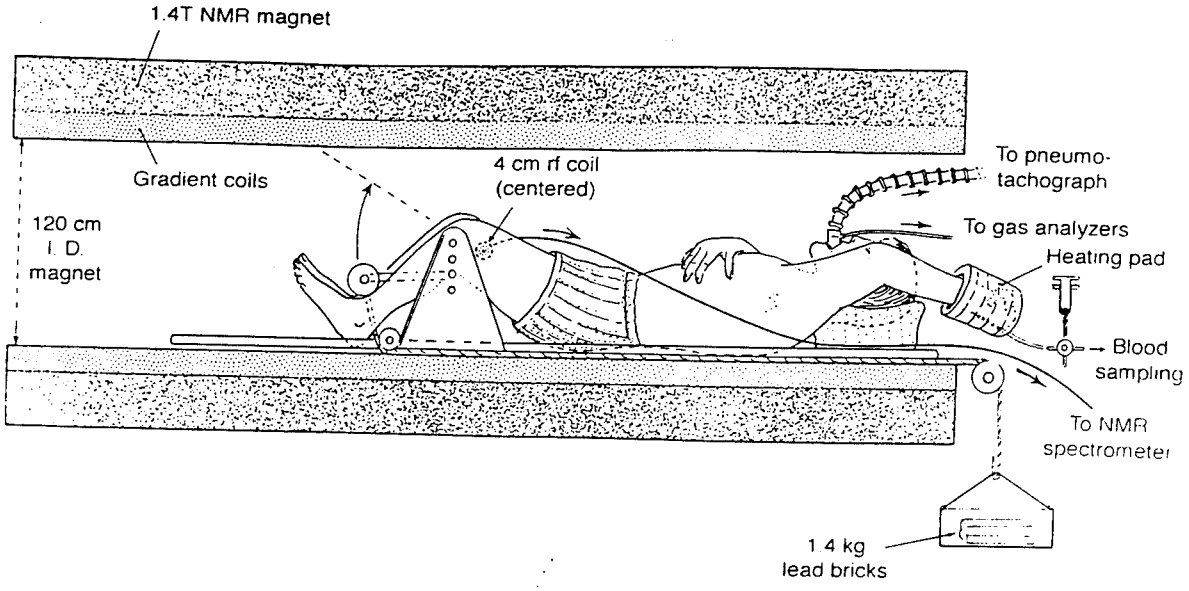

Fig. 1. Experimental setup for simultaneous ^{31}P-magnetic resonance spectroscopy (MRS) of vastus medialis, heated hand vein blood drawing (when used) and breath-by-breath measurements of ventilation and gas exchange during incremental exercise.

^{31}P-Magnetic Resonance Spectrometry. MRS spectra were acquired every 30 sec during the 5 min of rest and continuously throughout exercise. Subjects were positioned inside the 1.4 Tesla magnet with an operating frequency = 24.071 MHz for ^{31}P spectroscopy. A 4 cm diameter radiofrequency (rf) coil was fastened 7 to 8 cm proximal to the superior aspect of the patella overlying the anteromedial right thigh. Since the volume of tissue sampled by such a coil is cone-shaped to a depth of approximately 2 cm, the muscle group sampled was largely the vastus medialis. Spectra were acquired from the average of 12 rf pulses of 100 μs duration, with a TR=2.5 sec. Spectral parameters were chosen to optimize the signal-to-noise ratio; partial saturation was therefore accepted and reported metabolite ratios are approximations of concentration ratios of these metabolites. Five hundred twelve sample points were collected with a sweep width of 2.5 KHz.

Data Analysis. Breath-by-breath ventilatory and gas exchange data were averaged over contiguous 30 sec periods since this was the temporal limit of resolution dictated by MRS signal-to-noise ratio. Resting measurements were taken when values for end tidal PCO_2, oxygen consumption ($\dot{V}O_2$), CO_2 output ($\dot{V}CO_2$) and $\dot{V}E$ varied by <10% over at least two continuous min. Peak oxygen uptake ($\dot{V}O_2$peak) was defined as the highest $\dot{V}O_2$ measured during the last min of exercise. The ventilatory threshold (VT) was defined as the $\dot{V}O_2$ which marked the onset of a sustained rise in the $\dot{V}E/\dot{V}O_2$ without a rise in $\dot{V}E/\dot{V}CO_2$ (Systrom et al., 1990). Thresholds were chosen independently by three experienced observers without knowledge of subject or dietary phase. There was agreement between at least two observers in all instances.

MRS spectra were analyzed by one investigator who was blinded to subjects' dietary phase. Intracellular pH (pH_i) was calculated using a calibration curve determined previously in our laboratory:

(1) $pH_i=6.85+\log(3.56-\partial)/(\partial-5.64)$

where ∂=chemical shift between the median area of phosphocreatine (PC) and P_i peaks. The pHi threshold (pH_iT) was defined as the $\dot{V}O_2$ which marked a sustained fall in pH_i greater than 0.07 pH units (the limit of resolution of our pH measurements) below the resting value.

PC/P_i was determined by relating computer-integrated areas under the respective ^{31}P spectroscopy peaks. PC/P_i vs. $\dot{V}O_2$ was described by a 2-straight line segment model (Vieth, 1989) of the rectilinear plot allowing determination of a breakpoint between rapid and slow change, the PC threshold (PCT).

All statistics were performed by StatView (Abacus Concepts, Berkeley, CA). Data were expressed as mean ± standard error of the mean (SEM), with n=9 unless otherwise stated. Differences at rest and during exercise between the HFAT and HCHO trials were compared using a paired Student's t-test or factorial ANOVA with a Fisher *post hoc* test. Continuous MRS data were compared to exercise time by simple linear regression. $P \leq 0.05$ was considered significant.

RESULTS

Dietary Analysis. Average daily energy and protein intake did not differ between the HFAT and HCHO trials (Table 1). For the group, average intake = 2647 ± 114 kcal (15 ± 0.1% protein, 10 ± 0.1% fat, 75 ± 0.2% CHO) for the HCHO and 2603 ± 113 kcal (16 ± 0.1% protein, 69 ± 0.2% fat, 15 ± 0.1% CHO) for the HFAT diets.

Table 1. Daily energy intake and composition during HCHO and HFAT.

	HIGH CARBOHYDRATE				HIGH FAT			
SUBJECT #	ENERGY (kcal)	PROTEIN (gm)	FAT (gm)	CHO (gm)	ENERGY (kcal)	PROTEIN (gm)	FAT (gm)	CHO (gm)
1	2116	79.7	25.6	392.9	2198	84.3	171.3	83.1
2	2837	104.7	32.3	537.0	2952	113.7	228.2	110.7
3	2196	82.3	26.7	407.9	2198	84.3	171.3	83.1
4	2643	98.6	29.7	500.7	2349	94.3	178.1	94.2
5	2992	112.9	34.6	568.4	2943	114.1	227.1	111.1
6	2365	90.9	26.6	445.9	2332	90.7	179.6	89.5
7	2852	107.1	32.5	543.2	2879	113.0	220.2	111.0
8	2791	106.5	31.9	530.8	2599	100.6	201.7	99.3
9	3028	117.4	34.5	574.3	2974	116.2	228.8	113.7
mean	2647	100.0	30.5	500.1	2603	101.2	200.7*	99.5*
SEM	114	4.4	1.2	22.8	113	4.4	8.6	4.2
% kcal		15	10	75.0		16*	69*	15*
SEM		0.1	0.1	0.2		0.1	0.2	0.1

n=9 normal volunteers. *p<0.05, HFAT vs HCHO.

For the group, there were no significant differences in body weight comparing Day 1 to Day 5 of each diet (HCHO = -0.34 ± 0.14, HFAT = -0.66 ± 0.21 kg). Five individuals, however, had weight loss ≥1% of their initial body weight (-1.13 ± 0.09 kg) after 5 days of the HFAT diet; only one experienced this degree of weight loss (-1.60 kg) with the HCHO diet. During the HFAT trial, 4 subjects reported gastrointestinal discomfort including fullness, nausea, abdominal cramping, gas, and diarrhea; one complained of mild intestinal gas during the HCHO diet.

Metabolic data. Collection of metabolic and MRS data began 3.8 ± 0.2 hr after HCHO and HFAT liquid meals, respectively. Both resting and peak exercise respiratory exchange ratio and $\dot{V}E$ were higher after HCHO (Table 2, Fig. 2).

Exercise time was greater in the HCHO (339 ± 34 vs. 308 ± 25 sec, HCHO vs. HFAT), but $\dot{V}O_2$peak was not (908 ± 131 vs. 873 ± 140 cc/min, HCHO vs. HFAT, Table 2, Fig. 2). The VT did not change, whether expressed as absolute $\dot{V}O_2$ or percent of $\dot{V}O_2$peak (n=6). Three subjects had no perceptible VT, however, following HFAT.

^{31}P Magnetic Resonance Spectroscopy. ^{31}P-MRS spectra from a representative control subject at rest and throughout exercise are shown in Fig. 3. For the group at rest, skeletal muscle PC/P_i and pH_i did not change as a function of diet (Table 2).

The two-segment rectilinear fit of PC/P_i vs. $\dot{V}O_2$ is shown for a representative individual on each of the two study days in Fig. 4. The slope of the initial steep fall of PC/P_i vs. $\dot{V}O_2$ was blunted for this and for 6 of 8 additional subjects when HCHO was compared to HFAT. Thus, for the group (Fig. 5), the slope of the steep phase was blunted ($-4.1 \cdot 10^{-2} \pm 0.7 \cdot 10^{-2}$ min/cc) following HCHO vs. ($-5.6 \cdot 10^{-2} \pm 1.2 \cdot 10^{-2}$ min/cc) after HFAT. The PCT was not affected by diet. The post-PCT slope of PC/P_i vs. $\dot{V}O_2$ was blunted following HCHO vs. HFAT in 6 of the 9 subjects. For the group, there was a tendency for the post-PCT PC/P_i slope to be blunted after HCHO ($5.77 \cdot 10^{-3} \pm 1.05 \cdot 10^{-3}$ min/cc vs. $6.35 \cdot 10^{-3} \pm 0.71 \cdot 10^{-3}$ min/cc, HCHO vs. HFAT; p=0.25). The PC/P_i at exhaustion was not influenced by diet (Fig. 6).

Table 2. Metabolic and spectroscopic measurements at rest and peak exercise after HCHO and HFAT.

	HIGH CARBOHYDRATE (mean)	(SEM)	HIGH FAT (mean)	(SEM)
REST				
VO2 (cc/min)	183	17	193	18
VO2 (cc/kg/min)	2.8	0.2	2.9	0.2
VCO2 (cc/min)	149	14	136	13
RQ	0.81	0.03	0.7*	0.02
VE (l/min)	7.8	0.6	7.3*	0.6
PC/Pi	9.8	0.4	9.8	0.3
pHi (units)	7.13	0.01	7.12	0.01
PEAK EXERCISE				
Exercise Duration (s)	339	34	308*	25
VO2 (cc/min)	908	131	873	140
VO2 (cc/kg/min)	13.6	1.8	13.0	1.8
VCO2 (cc/min)	1088	189	1008	204
RER	1.22	0.07	1.11*	0.07
VE (l/min)	41.3	7.7	36.8*	7.0
PC/Pi	0.61	0.20	0.61	0.19
pHi (units)	6.68	0.06	6.55	0.10

n=9 normal volunteers. *$p<0.05$, HFAT vs HCHO.

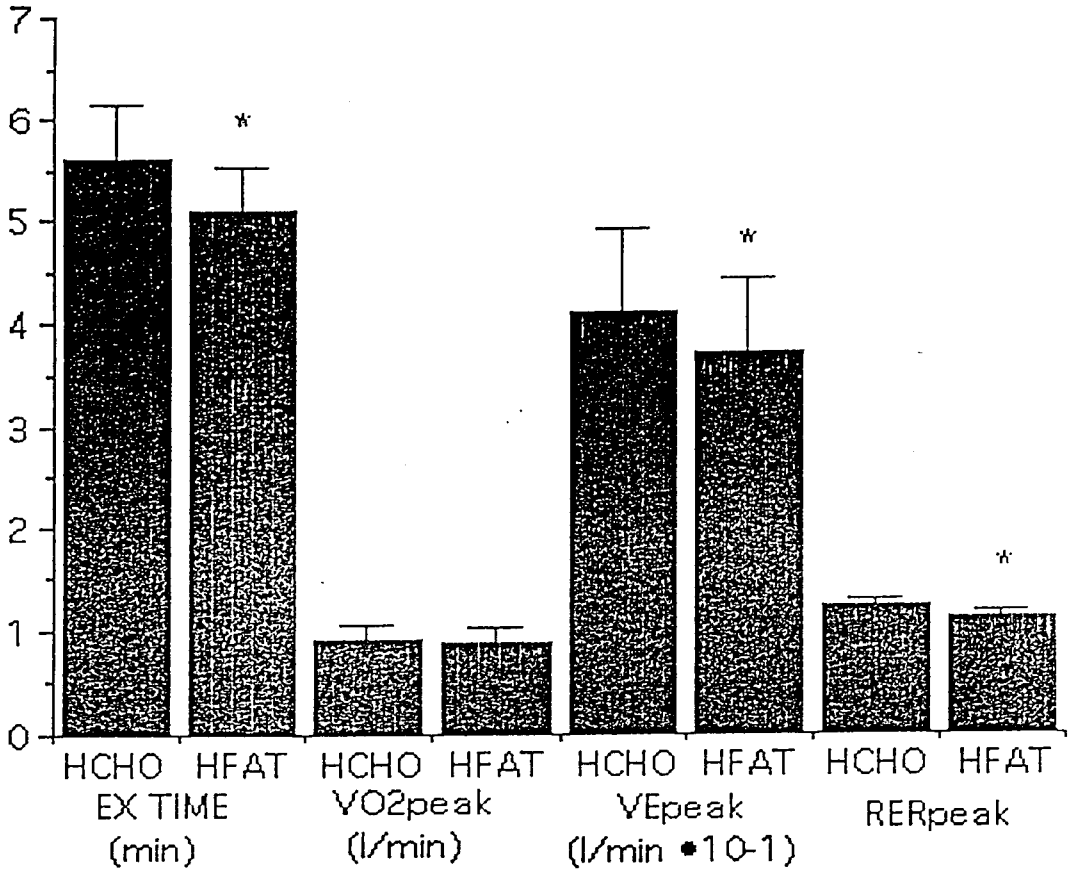

Fig. 2. Incremental exercise time, peak exercise ventilation and gas exchange following HCHO and HFAT. Date = mean ± SEM. *p<0.05.

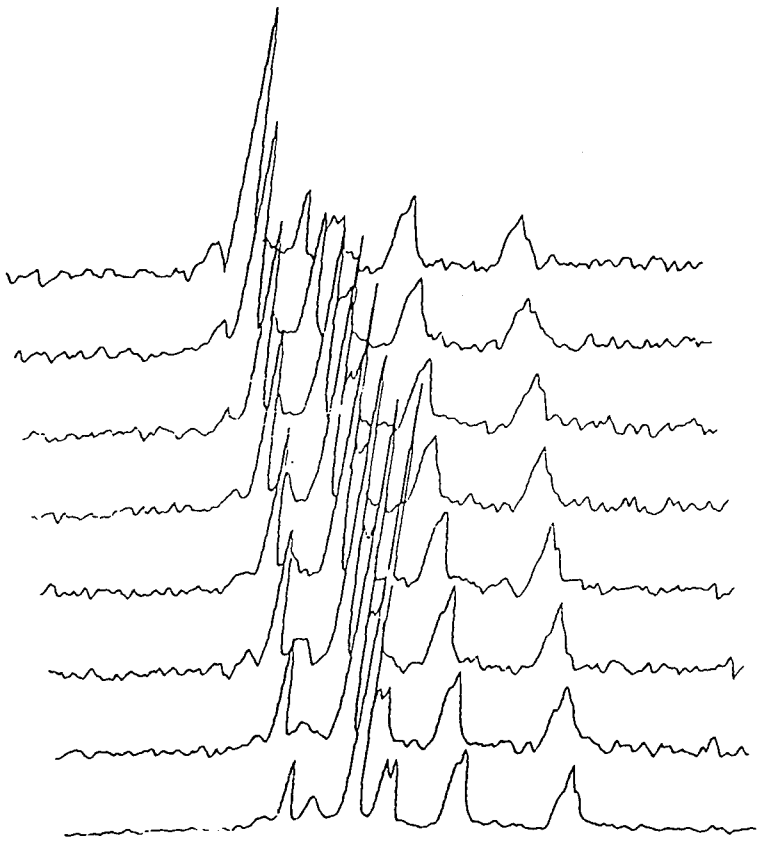

Fig. 3. ^{31}P-magnetic resonance spectra from vastus medialis at rest (foreground) and every 30 sec throughout incremental exercise to exhaustion in a representative normal individual.

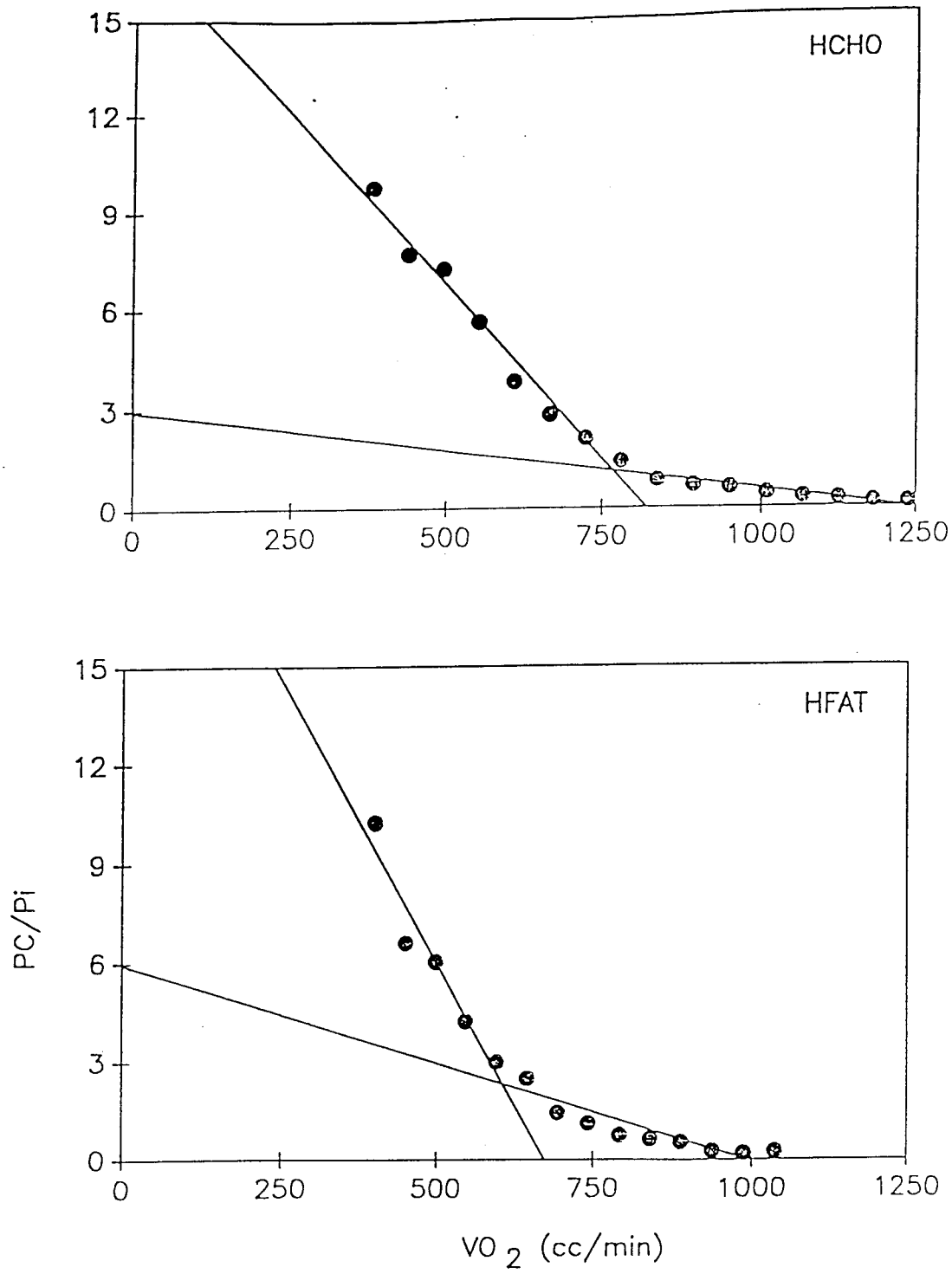

Fig. 4. Two-segment model of rectilinear plot of PC/P_i vs. $\dot{V}O_2$ for a representative individual during graded quadriceps exercise (see text). The $\dot{V}O_2$ at the intersection of the two segments denotes the PC threshold. After HCHO (upper panel), both pre and post PCT slopes are more shallow than after HFAT (lower panel).

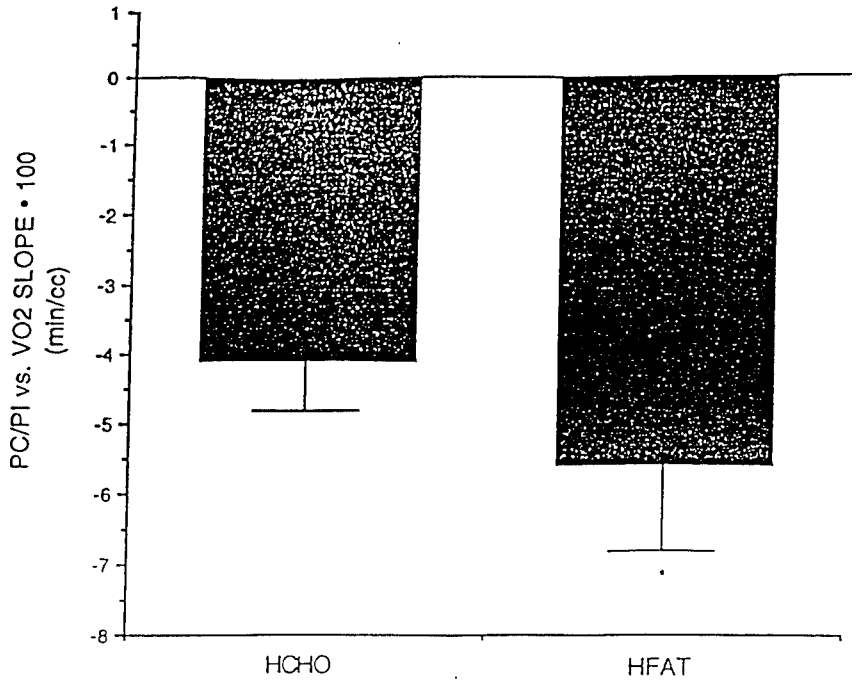

Fig. 5. For the groups, the steep (Pre PCT) slope of PC/P_i vs. $\dot{V}O_2$ is blunted following HCHO. Data = mean ± SEM. *$p<0.05$. Slope of PC/P_i vs. $\dot{V}O_2$ is a unitless term divided by cc/min to yield min/cc.

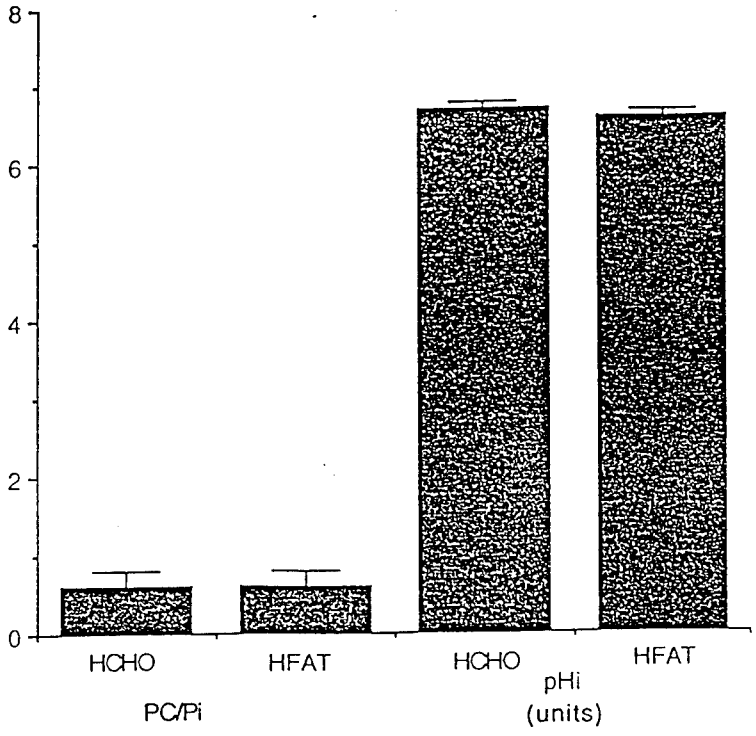

Fig. 6. MRS data at exhaustion. Diet had no effect on PC/P_i of pH_i during the last full 30 sec of incremental exercise. Data = mean ± SEM.

Irrespective of diet, exercise time was related to the slopes of PC/P_i vs. $\dot{V}O_2$ before and after the PCT (Fig. 7). The more shallow the slope before and after the PCT, the more prolonged the incremental exercise bout.

The pH_iT was not affected by diet whether measured as absolute $\dot{V}O_2$ (451 ± 53.7 vs. 480 ± 52.6, HCHO vs. HFAT; p=0.16) or percent of $\dot{V}O_2$peak (52 ± 3.5 vs. 60 ± 4.8%, HCHO vs. HFAT; p=0.07). The slope of pH_i vs. $\dot{V}O_2$ after the pH_iT was likewise unaffected ($-1.07 \cdot 10^{-3} \pm 0.13 \cdot 10^{-3}$ vs. $-1.49 \cdot 10^{-3} \pm 0.29 \cdot 10^{-3}$ units/cc/min; HCHO vs. HFAT; p=0.13). When pH_i on the two study days was compared at each decade of percent of $\dot{V}O_2$peak, no differences were seen by ANOVA (Fig. 8). Peak exercise pH_i tended to be slightly more alkaline following HCHO (6.68 ± 0.06 vs. 6.55 ± 0.09, HCHO vs. HFAT; p=0.09, Fig. 6).

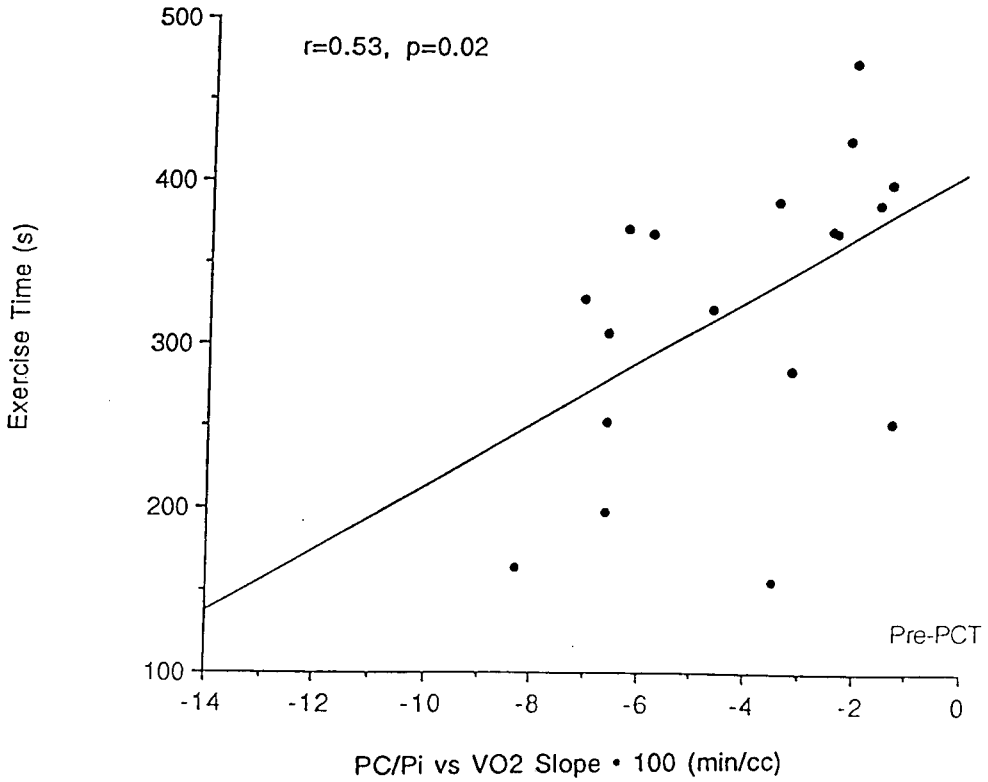

Fig. 7. Incremental exercise time was directly related to the slope of PC/P_i vs. $\dot{V}O_2$, before (upper panel) and after (lower panel) the PCT (see Discussion).

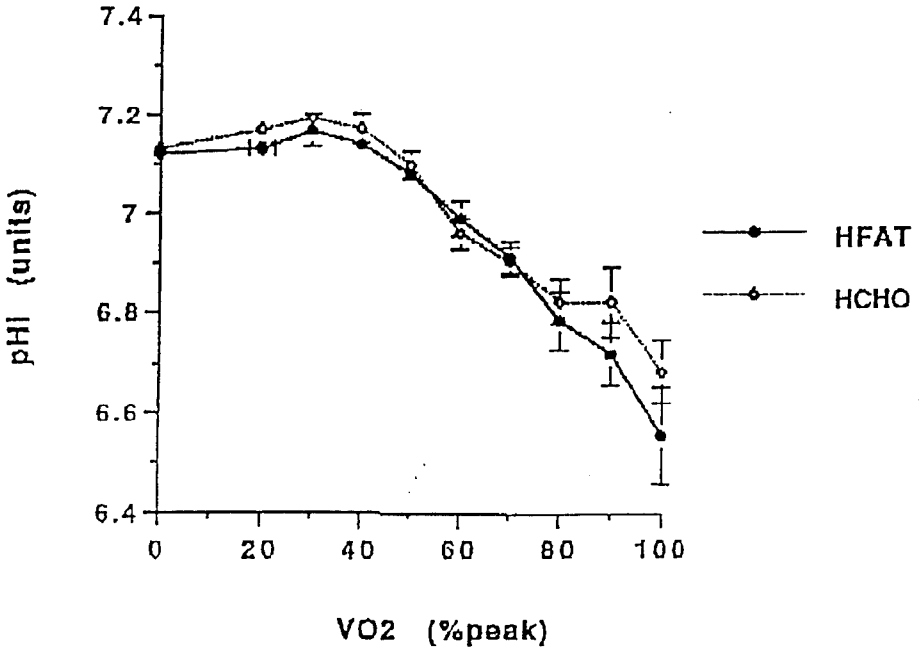

Fig. 8. Quadriceps pH_i at rest (0) and throughout incremental exercise as a percent of $\dot{V}O_2peak$, grouped by decade. By factorial ANOVA, diet did not influence pH_i at any metabolic rate. Data = mean ± SEM.

DISCUSSION

The principal findings of this study were that compared to a high-fat diet, a diet rich in CHO prolongs incremental exercise time, without affecting $\dot{V}O_2peak$ and the improved exercise performance is related to relative preservation of skeletal muscle PC/P_i. The data thereby confirm classic associations between diet and exercise performance and offer new insight into the cellular mechanisms linking the two.

Diet and substrate utilization. The current gas exchange data support the notion that antecedent diet influences exercising muscle's choice of substrate for energy production. This concept has been best established for endurance exercise following a dietary modification of several days' duration (Bergstrom et al., 1967; Christensen & Hansen, 1939; Costill et al., 1977). Similar conclusions have been reached for short-term, intense exercise (Greenhaff et al., 1988; Greenhaff et al., 1987; Maugham & Poole, 1981; McLennan & Gass, 1989; WhiteWilson et al., 1988; Yoshida, 1984). Several explanations have been offered to account for these findings. Liver and skeletal muscle glycogen stores rise after a CHO diet (Bergstrom et al., 1967; Coyle et al., 1985; Hermansen et al., 1967; Hultman & Bergstrom, 1967; Hultman et al., 1967; Jansson & Kaijser, 1982; Nilsson & Hultman, 1973) and presumably make CHO more readily available. Conversely, for at least 4 hr following ingestion of CHO, increased plasma insulin (or its lingering effects) suppresses lipolysis and free fatty acid (FFA) availability during subsequent exercise (Coyle et al., 1977).

In a similar fashion, fat seems to be preferentially oxidized by exercising muscle under conditions of decreased glycogen reserves (Bergstrom et al., 1967; Christensen & Hansen, 1939; Costill et al., 1977) and/or increased FFA availability (Costill et al., 1977; Jansson & Kaijser, 1982). Postulated mechanisms include FFA inhibition of muscle glucose uptake (Rennie & Holloszy, 1977), citrate-mediated inhibition of CHO oxidation (Jansson & Kaijser, 1982) and over the long-term, up-regulation of regulatory enzymes involved in FFA oxidation (Miller et al., 1984). Although best demonstrated for prolonged exercise, it appears that mobilization of muscle triacylglycerol can be important during short-term exercise as well (Jones et al., 1980).

Diet and exercise performance. The current data are also in accord with those who have demonstrated increased time to exhaustion after dietary CHO vs. fat, whether exercise was submaximal and prolonged (Bergstrom et al., 1967; Christensen & Hansen, 1939; Hermansen et al., 1967; Hultman & Bergstrom, 1967) or relatively maximal and brief (Greenhaff et al., 1988; Greenhaff et al., 1987, Maughan & Poole, 1981; McLellan & Gass, 1989; White et al., 1988; Yoshida, 1984). The findings are not universal, however, as Simi et al. found that a high-fat diet

marginally improved $\dot{V}O_2max$ in rats (Simi et al., 1991). However, in that study, the increased fat intake was significantly longer than in our study.

Diet, skeletal muscle metabolism, and exercise performance. Peripheral fatigue during near-maximal exercise is likely related to some combination of accumulation of toxic metabolites and depletion of energy stores (MacLaren et al., 1989). If and how manipulation of diet influences such "fatigue factors" is not completely understood.

Accumulation hypothesis. "Toxic" metabolites which may play a role in fatigue during brief, intense exercise include H^+, diprotonated phosphoric acid (Wilson et al., 1988), ADP, and P_i (Cooke & Pate, 1985). Skeletal muscle acidosis during short-term exercise is largely due to narrowing of the strong-ion difference, in turn secondary to an imbalance between lactic acid production on the one hand and its buffering, oxidation, and membrane transport on the other (Kowalchuck et al., 1988). H^+ may limit exercise tolerance through changes in sarcolemmal potential, excitation-contraction coupling, and by inhibition of actomyosin cross-bridge formation (Maclaren et al., 1989). In the current study, no diet-related difference pH_i was found at rest or exercise as function of diet. The current data therefore do not support the conclusions of Greenhaff et al., who postulated that reduced high-intensity exercise time after a high-fat diet might be related to an observed mild relative resting metabolic acidemia and decreased buffering capacity (Greenhaff et al., 1988; Greenhaff et al., 1987). Rather, our data are in accord with those of Bertocci et al., who suggest that intracellular acidosis is not a prerequisite for fatigue during short-term exercise (Bertocci et al., 1992).

By ^{31}P-MRS, the peak resonance frequency for P_i moves toward PC as a function of decreasing pH_i and increasing diprotonated P_i. Our data are therefore compatible with those of Wilson et al. and a role for diprotonated P_i as an ergolytic byproduct of increased metabolism (Wilson et al., 1988).

In the current study, incremental exercise time was best related to the PC/P_i ratio, which in turn, reflects the phosphorylation potential of the cell (adenosine triphosphate [ATP]/ADP+P_i).

Another possible interpretation is that ADP and P_i, higher after increased dietary fat, directly inhibit actomyosin cross-bridge function (Cooke & Pate, 1985).

Exhaustion hypothesis. Fatigue during prolonged submaximal exercise has been well-related to muscle glycogen depletion (Bergstrom et al., 1967; Hermansen et al., 1967; Hultman, 1989; Jansson & Kaijser, 1984). During aerobic exercise, there is a rapid expansion of tricarboxylic acid (TCA) cycle intermediates (Sahlin et al., 1990) which is dependent on CHO availability (Spencer et al., 1991). Although such factors might be expected to play a relatively minor role in short-term exercise, selective glycogen depletion has been shown in fast fatiguable fibers after brief, intense exercise (Gollnick et al., 1974). Thus, it is possible that reduced CHO availability due either to depletion of glycogen stores or increased fat intake can compromise short-term exercise capacity. The precise nature of the defect in energy transduction under these conditions remains unclear, however.

In the current study, improvement in incremental exercise time after a HCHO diet was related to relative preservation of PC/P_i vs. $\dot{V}O_2$. In a classic muscle biopsy study, Hultman et al. measured lower PC at several workloads after glycogen depletion (Hultman et al., 1967). Using phosphate MRS, Pichard et al. found decreased PC/P_i in resting muscle after both an hypocaloric diet and 2-day fast (Pichard et al., 1988). Koruda et al. found depressed skeletal muscle PC/P_i during simulated exercise in rats following a fast of 6-day durations (Koruda et al., 1985). Most recently, Bertocci et al. were able to relate fatigue to decreased PC and increased P_i, in both control experiments and when prior exercise had attenuated glycogen stores (Bertocci et al., 1992). Finally, patients with muscle phosphorylase deficiency, who rely nearly exclusively on FFA for energy production, exhibit fatigue associated with rapid and profound reduction in muscle PC/P_i at low metabolic rates (Lewis et al., 1985). Intravenous glucose partially reverses both the abnormal intracellular bioenergetic state and exercise capacity in such patients (Lewis et al., 1985). Thus, the current data and those of others suggest relative unavailability of glucosyl equivalents begets fatigue characterized by an inordinate fall in skeletal muscle PC/P_i.

PC represents a high energy "buffer" (Meyer et al., 1984) and under conditions of increased actomyosin ATPase activity, decreases in PC/P_i reflect a stressed cellular bioenergetic state. In exercising man, ATP is synthesized more rapidly when CHO (~2.4 and 1.0 mol/min, anaerobically and aerobically, respectively, for 28 kg of muscle) is oxidized vs. fat (~0.5 mol/min) (Hultman, 1989). Thus, with rapidly increasing ATP demand during incremental exercise one would expect a relatively preserved PC/P_i ratio if CHO is associated with more rapid high-energy phosphate production.

We, as others (McLellan & Gass, 1989; Yoshida, 1984), found evidence for improved short-term exercise "efficiency" (exercise time/$\dot{V}O_2$) after HCHO. Does the blunted fall in skeletal muscle PC/P_i after CHO offer any mechanistic insights? The high-energy phosphate regeneration (P) to $\dot{V}O_2$ ratio is 4% to 11% higher when CHO rather than FFA is oxidized ($P:O_2$=5.9-6.3 for CHO; 5.60-5.65 for FFA (McGilvery, 1983). Relative preservation of PC/P_i after CHO may therefore reflect more "potent" ATP production when CHO rather than fat is the oxidized substrate of choice.

The conclusion that CHO's beneficial effects on exercise performance are mediated by improved mitochondrial function and phosphorylation potential requires that the observed PC/P_i changes were not mediated by pH_i and the creatine kinase reaction. By mass action, a relatively alkaline pH_i throughout incremental exercise after HCHO could have inhibited PC splitting and preserved PC/P_i. We did not find changes in pH_i as a function of diet at any percent of $\dot{V}O_2$peak, though a trend was apparent at exhaustion. Even if this finding at $\dot{V}O_2$peak were significant, it could not explain relative increases in PC/P_i throughout submaximal exercise, however.

Limitations of the study. For budgetary reasons we did not hospitalize our volunteers for direct observation and administration of the prescribed dietary regimens. Significantly higher respiration exchange ration after HCHO vs. HFAT suggested compliance with diet, however. To avoid (de)training effects, subjects' exercise habits were not changed during the dietary manipulation phase. Since subjects' normal exercise habits ranged from sedentary to regular, it is possible the protocol induced different degrees of glycogen depletion during HFAT. Since

muscle glycogen was not measured, it is difficult to determine if observed differences were due to changes in diet *per se*, glycogen reserves, or both.

CONCLUSIONS

It is concluded that diet influences exercise performance at least in part through changes in skeletal muscle high-energy phosphates. Carbohydrate likely improves short-term incremental exercise efficiency because of its inherent ability to rapidly generate more energy per mole of O_2 consumed.

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To determine how diet modulates short-term exercise capacity, skeletal muscle pH, and bioenergetic state were examined by ^{31}P-magnetic resonance spectroscopy (MRS) in nine healthy volunteers. Subjects performed incremental quadriceps exercise to exhaustion following 5 day of high-CHO (HCHO) or high-fat (HFAT) diet which were randomly assigned in cross-over fashion and separated by a 2.5 day period of ad libitum mixed diet. Simultaneous measurements were made of pulmonary gas exchange, minute ventilation, and quadriceps muscle pH and phosphorylation potential. At rest and peak exercise, respiratory exchange ratio and $\dot{V}E$ were higher following HCHO vs. HFAT ($p<0.05$) reflecting greater CHO utilization. $\dot{V}O_2$peak was not increased after HCHO ($p>0.05$), but exercise duration was (339±34 vs. 308±25 sec, HCHO vs. HFAT; $p<0.05$). HCHO was associated with a blunted early fall of PC/P_i vs. $\dot{V}O_2$ ($-4.1\cdot10^{-2}\pm0.7\cdot10^{-2}$· vs. $-5.6\cdot10^{-2} \pm 1.2\cdot10^{-2}$ min/cc, HCHO vs. HFAT; $p<0.05$). On both study days, the slope of PC/P_i vs. $\dot{V}O_2$, before and after the PC threshold, was correlated with exercise time. The results suggest that a diet rich in CHO improves exercise efficiency through beneficial effects on intracellular phosphorylation potential.

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