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Article *in* Medicine & Science in Sports & Exercise · October 1993
DOI: 10.1249/00005768-199309000-00007 · Source: PubMed

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Carbohydrate dependence during marathon running

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ABSTRACT

O'BRIEN, M. J., C. A. VIGUIE, R. S. MAZZEO, and G. A. BROOKS. Carbohydrate dependence during marathon running. *Med. Sci. Sports Exerc.*, Vol. 25, No. 9, pp. 1009–1017, 1993. To test the hypothesis that marathon running is dependent on lipid oxidation, 12 post-absorptive males (31.9 ± 2.1 yr) ran a treadmill marathon and substrate utilization was assessed. Subjects were placed into a fast (F ≤ 2 hr, 45 min; 73.3% $\dot{V}O_{2max}$), or a slow (S ≤ 3 hr, 45 min; 64.5% $\dot{V}O_{2max}$) marathon group. The day before testing subjects rested, but ate their normal diet. Subjects were tested in the morning after an overnight fast, and only tap water, at a rate of 1 l/h, was ingested during exercise. Blood glucose concentration rose at exercise onset, peaked at approximately an hour, but then decreased over time remaining at or above resting levels. Free fatty acids and glycerol rose continuously. No significant differences in plasma FFA, glycerol, or blood glucose concentrations were observed between F or S groups during the marathon. Mean blood lactate concentration was significantly higher ($P < 0.05$) in the F (2.1 ± 0.3 mM) group than the S (1.2 ± 0.2 mM) during exercise. Mean plasma epinephrine was significantly higher in the F (0.9 ± 0.2 ng·ml^{-1}) than the S (0.6 ± 0.2 ng·ml^{-1}) group; norepinephrine was also higher in F (3.9 ± 1.4 ng·ml^{-1}) than the S (2.5 ± 0.9 ng·ml^{-1}, $P \leq 0.05$). Blood lactate and epinephrine concentrations correlated significantly (r = 0.76 and 0.78 in F and S groups, respectively). The average respiratory gas exchange ratio (R = $\dot{V}CO_2/\dot{V}O_2$) was higher in F (0.99 ± 0.01) than S (0.90 ± 0.01, $P \leq 0.05$). A direct relationship between carbohydrate oxidation and running speed during marathon running is indicated. Estimated carbohydrate combustion [(F: 2,414.3 ± 43.0 kcal (575 ± 10 g); S: 2,890.0 ± 159.0 kcal (688 ± 38 g)] exceeded estimated glycogen stores in active muscle and liver (475 g = 375 g (muscle) + 100 g (liver)]. Therefore, total body glycogen stores were made available for combustion. All classes of energy substrates participate, but carbohydrate, not lipid, is the primary fuel for marathon running.

GLUCOSE, LACTATE, FATTY ACIDS, EPINEPHRINE, INSULIN, ENERGY EXPENDITURE, LACTATE SHUTTLE, GLUCOSE FATTY ACID CYCLE, EXERTION

Endurance training, such as that practiced for marathon running, results in a doubling of muscle mitochondrial content (16,17,21) through an elaboration of the mitochondrial reticulum (28). This adaptation leads to a greater capability to oxidize all fuel substrates including carbohydrates (CHO), lipids [free fatty acids (FFA) and intramuscular triglycerides (TG)] (34), and amino acids (5). The increased capacity to oxidize fatty acids coinciding with mitochondrial adaptation is thought to result in a greater reliance of lipid over CHO fuel sources during exercise (21,34), thereby sparing muscle glycogen and allowing the limited glycogen reserves available in active muscle to sustain prolonged exercise. The conclusion that endurance training results in a shift toward lipid dependence during sustained exercise is based on lower respiratory gas exchanges ratio (R = $\dot{V}CO_2/\dot{V}O_2$), blood lactate level, and net muscle glycogenolysis at given absolute work loads before and after training (17). However, increased FFA utilization in trained compared with untrained individuals during marathon running or other forms of sustained high intensity exercise has not been demonstrated. In fact, using isotopic tracers, Hall et al. (19) observed blood glucose turnover to increase approximately threefold during marathon running, whereas FFA (palmitate) turnover increased only slightly. In another report Jones et al. (26) found blood palmitate turnover to decline during sustained high intensity (70% $\dot{V}O_{2max}$) exercise compared with rest.

Contrary to the widely held opinion that marathoners rely predominantly on lipid fuel sources, there is evidence that can be interpreted to suggest reliance on body carbohydrate stores during sustained exercise requiring high power outputs as in marathon running. It is known that arterial glucose concentration and rate of appearance are maintained higher in trained than in untrained rats (38) and men (29) during exercise. Further, it is known that insulin action (27) and skeletal muscle hexokinase activity (3) are increased by training. Thus, it is possible that the apparent glycogen sparing effect of endurance training is due to enhanced capacity

0195-9131/93/2509-1009$3.00/0
MEDICINE AND SCIENCE IN SPORTS AND EXERCISE

Submitted for publication April 1992.
Accepted for publication March 1993.

to utilize blood glucose as a fuel. In fact, Donovan and Sumida (15) have estimated that most of the glycogen-sparing effect of endurance training in exercising rats is due to increased glucose turnover. Increased gluconeogenesis in trained rats during exercise could only be explained by gluconeogenesis from lactate (9,15,38).

The limited data available on marathoners suggest carbohydrate dependence. Scrimgeour et al. (36) reported that in elite marathoners' gas exchange R values were directly related to speed of running and were close to 1.0 at race pace. Dependence on carbohydrate combustion in marathon runners was suggested. These results have recently been replicated by Bosch et al. (4) on 17 athletes who completed a treadmill marathon with an average gas exchange R of 0.93. Because active skeletal muscle lacks sufficient glycogen stores to sustain a marathon, how then is a marathon sustained by carbohydrate oxidation?

It was the purpose of the present study to investigate substrate utilization during marathon running at two different paces. The hypothesis tested was that the primary substrate would be fat in both fast and slow marathon groups. Alternatively, we also evaluated the hypothesis that carbohydrate is the primary energy source during marathon running with a greater rate of carbohydrate combustion in fast compared with slow marathon running. Our results show carbohydrate dependence during marathon running and implicate a significant role for the lactate shuttle (6) or alternative mechanism as a means of maintaining glycemia and supplying oxidizable carbohydrate.

METHODS

Subjects

Twelve nonsmoking trained male volunteers (Table 1) between the ages of 18–35 yr were assigned to fast (F) or slow (S) marathon running groups. Assignment to group was determined by sequence of recruitment. Subjects placed in the fast group were scheduled to complete the marathon distance in 2 hr 45 min whereas the slow group completed the marathon in 3 hr 45 min. Experimental procedures were approved by the University of California at Berkeley, Committee for the Protection of Human Subject (Protocol 86-10-15), and informed consent was obtained.

Exercise Protocols. Subjects performed two exercise trials, separated by 1–2 wk. The first trial was a continuous, progressive treadmill protocol to maximum to determine maximal oxygen consumption ($\dot{V}O_{2max}$). The second trial was a treadmill marathon on 0% grade.

TABLE 1. Physical characteristics of subjects (mean ± SEM).

N	Group	Age (yr)	Weight (kg)	$\dot{V}O_{2max}$ $(l \cdot min)^{-1}$	$\dot{V}O_{2max}$ $(ml \cdot kg^{-1} \cdot min^{-1})$
6	Slow (S)	29.9 ± 6.2	73.2 ± 10.1	4.02 ± 0.7	55.0 ± 7.3
6	Fast (F)	30.2 ± 5.2	66.8 ± 8.8	3.94 ± 0.3	58.9 ± 3.6
12	F and S	30.0 ± 5.5	70.3 ± 10.0	3.98 ± 0.48	56.7 ± 6.2

Graded Exercise to Maximum ($\dot{V}O_{2max}$)

Subjects rested for 10–15 min after several minutes of exercise to familiarize them with treadmill running. The protocol to evaluate $\dot{V}O_{2max}$ was a continuous incremental test, with the first stage being 3.0 miles $\cdot h^{-1}$, 2 1/2% grade on a Quinton (Q65) motorized treadmill. Thereafter, the speed and gradient increased every 2 min until voluntary cessation. Respiratory gas exchange ($\dot{V}O_2$, $\dot{V}CO_2$ and R) was measured on-line using standard open circuit techniques (7,30). Inspired ventilatory flow was measured with a Fleisch pneumotachometer (no. 3), Validyne MP45 pressure transducer, and DC15 carrier demodulator. Expired gas was sampled continuously from a 5-l mixing chamber and passed via a heated line through AMETEK oxygen (S-3A) and carbon dioxide (CD 3-A) analyzers. Analog signals from the three sensors were digitized in an IBM interactive board and IBM-AT computer was used for calculation of respiratory gas exchange parameters as well as for data storage and retrieval.

Marathon Running

Subjects reported to the laboratory in the morning after an overnight fast. On the day prior to the marathon, subjects rested and ate their usual diet in normal amounts. The overnight fast was intended to maximize lipid metabolism and control for possible effects of variations in the immediate, premarathon eating schedule. Prior to the marathon, an 18.5-gauge Teflon catheter was inserted into a superficial forearm vein for blood sampling. Catheter patency was maintained by intermittent flushing of nonheparinized physiological saline.

Measurement of respiratory gas exchange during marathon running was performed as during assessment of $\dot{V}O_{2max}$. In addition to routine on-line analyses of expired air, at random expired air samples were collected in mylar bags and glass syringes for analyses on a second set of electronic analyzers and by Scholander technique (10).

Blood samples were drawn at 10 min before exercise, 5, 15, 30, 45, 60, and every subsequent 30 min until the end of the marathon, as well as at the end of exercise, and 10 min during post-exercise recovery. Subjects were required to drink 500 ml of tap water 15 min prior to running and each half hour of running to prevent excessive body water loss. Air flow from a large electric fan was directed on subjects to maintain their level of comfort. Subjects watched movies displayed on a video at the front of the treadmill.

Chemical Analyses

Glucose and lactate. Aliquots of blood samples were immediately deproteinized in chilled 6% perchloric acid. These were centrifuged, and the supernatant decanted. Lactate concentrations were determined enzymatically on neutralized acid extracts according to Hohorst (20). Blood glucose concentrations were determined utilizing hexokinase (Sigma Diagnostic no. 16-UV).

Hormones. Plasma catecholamines concentrations were determined by HPLC as described previously (31). Briefly, plasma samples were stored with an antioxidant (sodium metabisulfite, 0.4 $mg \cdot ml^{-1}$). Samples were stored at −80° until analysis when 100 μl of sample eluant were injected into the HPLC column (reverse mobile phase, Bio-Sil ODS-5S, Bio-Rad) and eluted with mobile phase (6.8 g sodium acetate-anhydrous, 1.0 g sodium heptane sulfonate, 60 ml acetonitrile, 1.0 Na_2 EDTA in 1 liter, pH adjusted to 4.8). The flow rate was 1.4 $ml \cdot min^{-1}$ at 2,000 psi with a potential of 0.65 V. The chromatogram was recorded on a RYT recorder (Bioanalytical Systems).

Insulin levels in arterial plasma were determined by radioimmunoassay (18).

Plasma free fatty acids and glycerol. Plasma free fatty acid (FFA) concentrations were determined on fresh samples according to Dole and Meinertz (12). Glycerol levels were measured in deproteinized blood according to Wieland (39).

Energy Expenditure

Carbohydrate combustion was assessed on the assumption that the gas exchange R equaled the nonprotein RQ. Glycogen reserves in active muscle were estimated assuming the active muscle mass to be 15 kg and glycogen content to be 25 $g \cdot kg^{-1}$ muscle for a total of 375 g (22). Further, it was assumed that liver contained 100 g of glycogen (33). Thus, the total glycogen reserves in active muscle and liver were estimated to be 475 g, or the equivalent of 1,900 kcal.

Statistical Analyses

Means and standard errors of metabolic parameters at each time point were computed. Significance of changes in blood metabolites and hormone levels within trials was determined using analysis of variance (ANOVA) with Scheffé *post-hoc* analyses to identify differences. Representative values for each subject during rest and exercise were obtained by averaging values obtained under those conditions. Significance of differences between fast and slow groups during rest and exercise were determined using two-way ANOVA. Statistical analyses were performed using CRISP software (Crunch Inc., San Francisco). An alpha of 0.05 was used throughout.

RESULTS

The fast group finished the marathon in 163 ± 8 min whereas the slow (S) group finished in 210 ± 11 min. The fast group also ran at a significantly higher mean $\dot{V}O_2$ (44.5 $ml \cdot kg^{-1} \cdot min^{-1}$ vs 35.5 $ml \cdot kg^{-1} \cdot min^{-1}$). These represented 73.3 ± 3.2 and 65.0 ± 4.7% of $\dot{V}O_{2max}$, respectively. The fast group also ran at a significantly higher gas exchange R ($P \leq 0.05$) (Fig. 1). Gas exchange R values declined over time in the slow group and at the end of exercise approximated 0.84.

Blood glucose levels fell initially in the first 15 min in the fast group, but then rose until 60 min into exercise (Fig. 2) ($P \leq 0.05$). Thereafter, blood glucose concentration fell, but rebounded near the end of exercise. Blood glucose concentration increased significantly ($P \leq 0.05$) in the slow group until 40 min, but then steadily declined until the end of exercise. There were no significant differences in blood glucose concentration between F and S groups during marathon running. In both fast and slow groups, blood glucose remained at or above preexercise values. During recovery

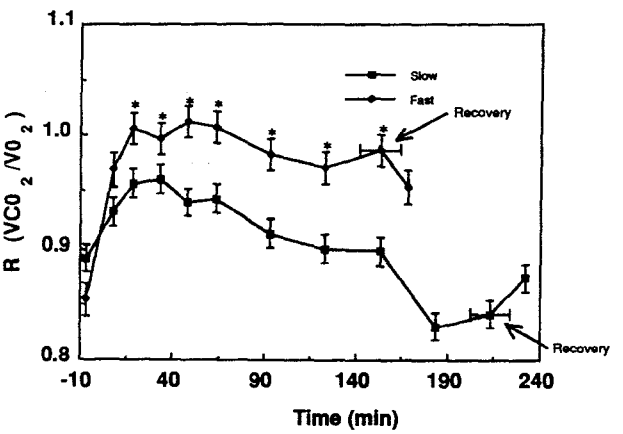

Figure 1—Respiratory exchange ratio ($R = \dot{V}CO_2/\dot{V}O_2$) for fast (F) and slow (S) groups during a treadmill marathon. Values are means ± SEM; $N = 6$ per group; asterisks indicate significant differences between groups.

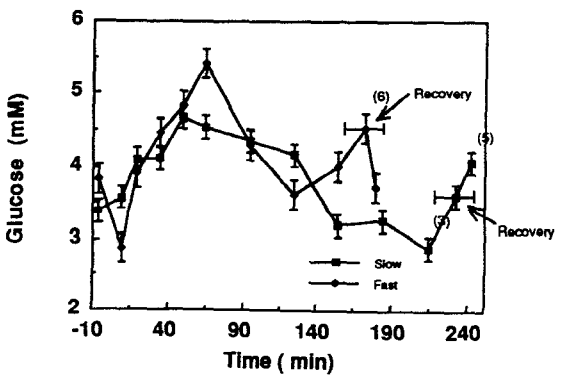

Figure 2—Blood glucose concentrations before, during, and 10 min after a treadmill marathon in fast (F) and slow (S) groups. Values are means ± SEM; $N = 6$ per group.

blood glucose levels increased in the slow group whereas glucose levels decreased in the fast group.

Blood lactate concentration in the fast group remained significantly higher than in the slow group throughout the marathon (Fig. 3). Within the slow group, lactate levels remained relatively stable throughout the marathon, with a slight increase after 120 min. During the short recovery period, blood lactate concentrations rose, particularly in the fast group.

Plasma FFA concentration rose continuously in both groups over the course of the marathon ($P \leq 0.05$) and peaked in recovery (Fig. 4). There were no statistically significant differences in blood FFA levels between groups during the marathon. Plasma glycerol mimicked plasma FFA levels and rose during running (Fig. 5). However, unlike FFA, plasma glycerol decreased during the recovery period in both groups. As with FFA, there were no significant differences in blood glycerol between the two groups throughout the exercise protocol.

Plasma epinephrine (E) levels were significantly elevated from preexercise values by 150 min in the fast group ($P \leq 0.05$). Additionally, plasma E in the fast group was significantly higher than slow group by 120 min of exercise ($P \leq 0.05$). In the slow group, epinephrine did not rise significantly until the end of exercise (Fig. 6). Blood lactate and plasma epinephrine values correlated significantly; r = 0.76 in the fast group, and r = 0.78 in the slow group.

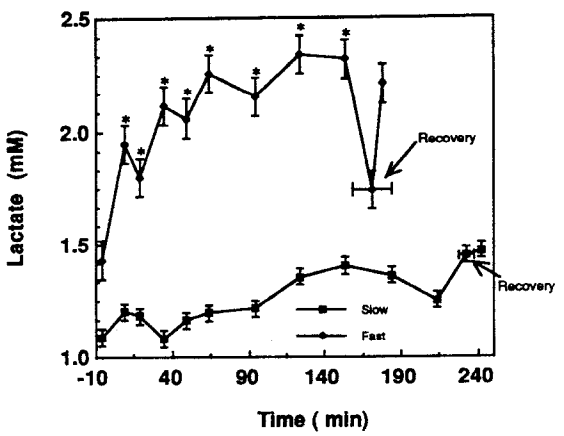

Figure 3—Blood lactate concentrations before, during, and 10 min after a treadmill marathon in fast (F) and slow (S) groups. Values are means ± SEM; $N = 6$ per group; asterisks indicate significant differences between groups.

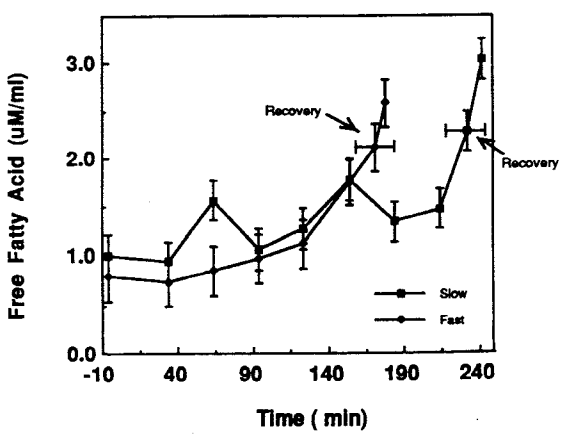

Figure 4—Plasma free fatty acid levels before, during, and after a treadmill marathon in fast (F) and slow (S) groups. Values are means ± SEM; $N = 6$ per group.

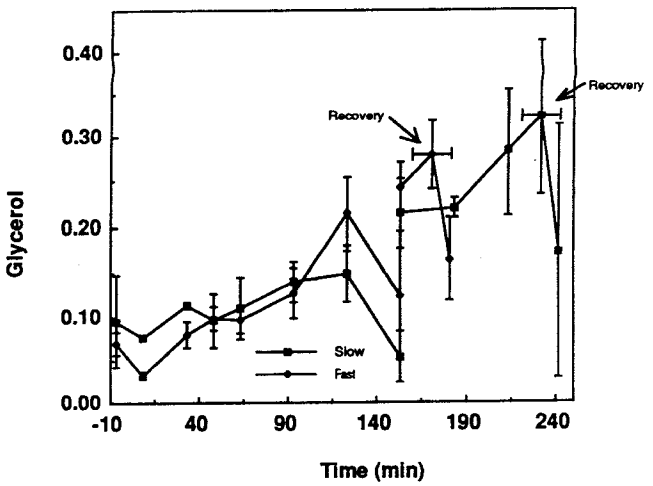

Figure 5—Plasma glycerol levels before, during, and 10 min after a treadmill marathon in fast (F) and slow (S) groups. Values are means ± SEM; $N = 6$ per group.

Plasma norepinephrine (NE) levels were elevated significantly in the fast group throughout the entire marathon ($P \leq 0.05$). Compared with the fast group, in the slow group NE levels did not rise as rapidly in the slow group during exercise. Plasma NE concentration fell at the end of exercise and during recovery in both groups. There were no significant differences in the NE/E ratios between fast or slow groups during the marathon (Fig. 6).

Plasma insulin levels declined in both the fast and slow groups during the marathon (Fig. 7). Although insulin tended to be lower in the fast group, there were no significant differences between the two groups in their insulin responses to prolonged submaximal exercise.

The percent of CHO combusted peaked at 60 min at 99% and 82% for the fast and slow groups, respectively (Fig. 8). Despite the declining percentage of CHO to overall energy expenditure from 60 min on, the fast group maintained higher percentage of CHO combustion than the slow group in which lipid apparently became the predominant fuel near the end of exercise. There was no significant difference between groups in the amount of total carbohydrate utilized; the higher rate of CHO oxidation in the fast group was compensated by a longer duration in the slow group such that the total CHO combustions were not different (Fig. 8). However, in both groups the amount of carbohydrates combusted [(F: 2,414.2 ± 43.0 kcal (575 ± 10 g); S:

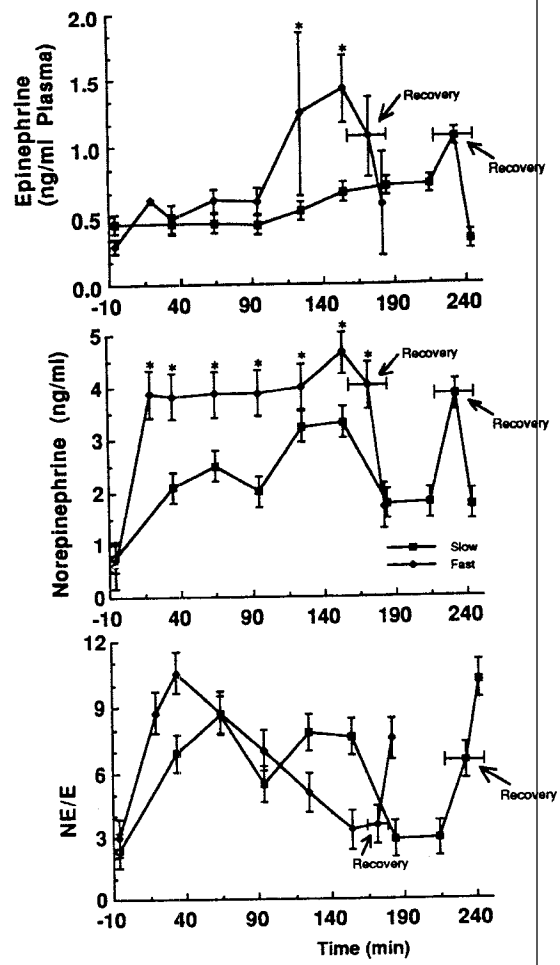

Figure 6—Plasma epinephrine, norepinephrine, and the ratio of the two catecholamines before, during, and 10 min after a treadmill marathon in fast (F) and slow (S) groups. Values are means ± SEM; $N = 6$ per group; asterisks indicate significant differences between groups.

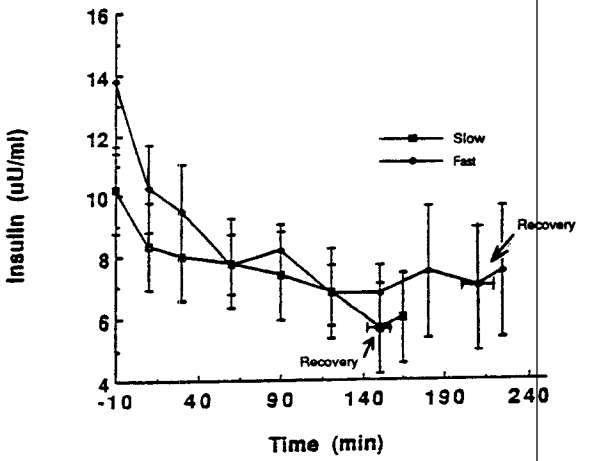

Figure 7—Plasma insulin concentrations before, during, and 10 min after a treadmill marathon in fast (F) and slow (S) groups. Values are means ± SEM. $N = 6$ per group; asterisks indicate significant differences between groups.

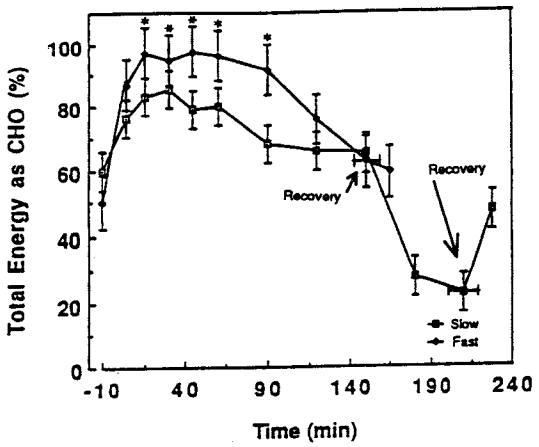

Figure 8—Calculated percentage of energy expenditure contributed by carbohydrates before, during, and after a treadmill marathon in fast (F) and slow (S) groups. Values are means ± SEM; $N = 6$ per group.

2,890.0 ± 159.0 kcal (688 ± 38 g)] exceed that estimated to be stored in the active muscle beds plus liver [475 g; 1,900 kcal].

DISCUSSION

Measurements of respiratory gas exchange and blood metabolite levels in experienced marathon runners cannot be interpreted to support the hypothesis that marathon runners are reliant on blood borne free fatty acids and intramuscular triglycerides as fuel sources. Instead, based upon our measurements of marathon $\dot{V}O_2$ and R, blood metabolite and hormone levels, and calculations of substrate utilization, we conclude that, in the mix of fuels used to accomplish a marathon, carbohydrates are the primary fuel source. Further, because carbohydrate oxidation exceeded estimated active muscle and liver glycogen stores, by deduction it must be that the ability to maintain carbohydrate combustion comes from an ability to mobilize carbohydrate stores from inactive muscle beds (2) and other tissues such as adipose (32) and skin (25). Although it is true that we lack data on blood FFA or muscle triglyceride (TG) turnover to support our conclusion in this regard, our data are consistent with those of Hall et al. (19) and Jones et al. (26), who determined that blood FFA turnover changed little or declined in sustained moderate to heavy intensity exercise. We believe that a careful reading of the literature shows our results, indicating carbohydrate dependence during marathon running to corroborate the results of others who have measured respiratory gas exchange during marathon running (Table 2).

Blood Metabolite Levels During a Marathon

Blood glucose rose initially in both fast and slow groups during the marathon, and then declined remain-

TABLE 2. Summary of published studies on respiratory gas exchange during marathon and long distance running.

Investigation	N	Group	Distance Run (miles)	Relative Effort (% $\dot{V}O_{2max}$)	Running Time (min)	R
Present study	6	Fast	26.2	73	165	0.99
	6	Slow	26.2	65	225	0.90
Bosch et al. (4)	17	Black and white	26.2	78.5	158	0.93
Scrimgeour et al. (36)	10	Training <60 km · wk^{-1}	Marathon pace	62	6	0.85
	10	Training 60–100 km	Marathon pace	62	6	0.89
	10	Training >100 km	Marathon pace	68	6	0.92
Costill (11)	1	Exhaustion	20.0 miles	69–78	200	0.93
Adams et al. (1)	1	Heat-acclimatized runners	26.2	—	165	0.87
Average						0.91

ing at or above preexercise levels (Fig. 2). In contrast, free fatty acid and blood glycerol levels rose continuously during marathon running (Figs. 4 and 5). Of all the blood metabolites measured, only lactate differed significantly between fast and slow groups. Because turnover rates of glucose, lactate, and free fatty acids are known to be correlated with arterial concentrations, the results do not give clear evidence of glucose-fatty acid cycle activity in which glucose utilization is suppressed by fatty acids during marathon running. Consistent with this conclusion are results of Hall et al. (19), who observed glucose turnover to increase during treadmill running, and Jones et al. (26), who observed blood palmitate turnover to decline in men exercising at 70% of $\dot{V}O_{2max}$.

Data from the present investigation demonstrate that blood lactate concentration increases significantly in experienced marathoners, especially runners exercising at 73% of $\dot{V}O_{2max}$ (Fig. 3). The higher lactate levels in the fast group shows further evidence that lactate production and accumulation are occurring in marathon runners at race pace. Although we have no data on the pathways of lactate disposal, it is likely that 75% of lactate was disposed of by direct oxidation (9,13,14,30,37) and the remainder to gluconeogenesis (7,8,37,38). The significant elevation in blood lactate concentration in the F group during marathon running could account for the increased rate of CHO oxidation in this group. In a recent investigation in which blood glucose and lactate fluxes were measured simultaneously in post-absorptive exercising men, it was apparent that most of the hepatic glucose production (glucose rate of appearance) came from lactate via the Cori cycle (7,8). However, because the fate of glucose during exercise is mainly oxidation (9), ultimately, most of the lactate formed during marathon running must be combusted.

Immediately in recovery, blood lactate rose in the fast marathon group (Fig. 3). This rise suggests that glycogenolysis and lactate production continued in recovery. Though catecholamines declined in recovery (Fig. 6), they remained above preexercise levels and likely contributed to the initial rise in blood lactate during recovery. The rise in blood lactate during recovery could also be explained by net glycogenolysis from inactive muscle beds (2).

In 1983, Donovan and Brooks (9,13) advanced the hypothesis that endurance training affects arterial lactate concentration during exercise mainly by enhancing clearance mechanisms. More recently, Donovan and Pagliassotti (14) have reported that when challenged with an exogenous lactate load, trained animals maintain lower blood concentrations because of enhanced capacities of lactate clearance via oxidation and gluconeogenesis with similar percentages of lactate disposed of via these two pathways. Results of these studies on animals are consistent with limited data on humans with different athletic histories (30,37). Trained and untrained humans during exercise demonstrate similar blood lactate appearance rates, but lower circulatory lactate levels in more highly trained individuals are attributable, in large part, to greater metabolic clearance rates. If, as suggested by results of our study, glycogen reserves in addition to those in liver and working muscle are mobilized by sympathoadrenal signals during marathon running, the distribution of carbohydrate potential energy in the form of lactate may provide the means by which glycogen reserves in diverse pools could be mobilized to support energetic requirements in active muscle.

Hormonal Levels During a Marathon

Results of the present investigation corroborate those of Hall et al. (19), who were the first to examine hormonal response in athletes during a marathon. For athletes fed a normal (45.5%) CHO diet for 3 d prior to study, Hall et al. observed catecholamines and T_3 to rise and insulin to fall during marathon running. In our study norepinephrine levels increased significantly in both fast and slow groups but obtained higher values in the fast group. Epinephrine increased in both groups by the end of the marathon. Whereas it is true that endurance training may dampen the catecholamine responses to given exercise power outputs (40), we reiterate that marathon athletes compete at relatively high power outputs (Table 2). That athletes train to compete at higher absolute as well as relative power

outputs has previously been made by Williams et al. (40). Thus, the impression of a dampened endocrine response to exercise in trained individuals is probably more imagined than real. Consistent with our interpretation, Kjaer et al. (29), using ^{3}H-epinephrine, observed an increased secretion of epinephrine in trained compared with untrained subjects despite lack of differences in blood glucose or heart rate between the two groups. In fact, Kjaer et al. have observed hyperglycemic and exaggerated glucose Ra, rate of appearance, responses in trained individuals during maximal exercise. The coordinated increases in blood glucose (Fig. 2) and norepinephrine we observed during marathon running (Fig. 6) are consistent with presence of a sympathetically mediated mechanism of gluco-regulation in marathoners. In this regard, Brooks et al. (7) have recently demonstrated a high correlation between blood glucose rate of appearance and circulating norepinephrine in human subjects resting and exercising at sea level and 4,300 m altitude.

In our study, we observed both elevated epinephrine and decreased insulin during marathon running. Previously, an increase in epinephrine has been associated with decreased insulin secretion (24). Mazzeo and Marshall (31) have demonstrated that the concentration of epinephrine was dependent on relative power output. Thus, the endocrine responses observed in the present investigation are consistent with those previously observed. The increased levels of circulating catecholamines could have aided in the mobilization of hepatic as well as other glycogen reserves.

Respiratory Exchange Values During a Marathon

The respiratory exchange ratio (R) is the simplest and most widely used method for the determination of fuel utilization. Application of the gas exchange R to studies of substrate oxidation requires at least three assumptions. These are that the R = nonprotein RQ, that acid-base status does not change, and that lipogenesis does not occur during marathon running. Although the assumption of no amino acid catabolism is not absolutely valid (5), it is likely that the error is not large (5–10%) and that both relative fat and CHO oxidations are slightly overestimated. Additionally, because the RQ for amino acid and protein oxidation (0.83) is closer to that of lipids (0.71) than carbohydrates (1.0), any significant utilization of amino acids as fuels would bias interpretation of the gas exchange data (Fig. 2) in favor of fats. Moreover, because training increases leucine oxidation during exercise (5), it is likely that we have overestimated fat utilization more in the F than S group. Nevertheless, because the gas exchange Rs were high in both F and S groups, the magnitude of errors in estimating fat oxidation is likely small. Regarding the second assumption, it is probable that whereas the blood acid-base status changed during the rest to exercise transition, during steady-state exercise when blood lactate concentration was constant (Fig. 3) acid-base status was probably also constant. Consequently, bicarbonate pool size probably remained constant during exercise. And finally, with regard to assumptions in the interpretation of the gas exchange R, it is likely that during marathon running catabolic, as opposed to anabolic, processes predominated. Therefore, it is unlikely that lipogenesis affected the gas exchange R. In summary on use of the gas exchange R in the present investigation, it is reasonable to assume that the R provided a fair estimate of fuel utilization.

In Figure 1 we report respiratory gas exchange data from on-line, real time analyses. However, analysis of aliquots of expired air by independent means yielded identical results to those shown. Therefore, we believe that the results shown accurately portray the metabolic responses of the subjects we studied. Similarly, we are confident that our estimates of carbohydrate oxidation (Fig. 8) are reasonable.

Because following training, when mitochondrial oxidative capacity is greater and blood lactate accumulation is reduced during given submaximal exercise power outputs, some investigators have concluded that there was a shift from carbohydrate to fatty acids in substrate utilization (17,21,34). In the main, the experimental paradigm used to reach this conclusion has involved measurement of gas exchange R during leg cycle ergometry at given absolute power outputs before and after training. Whereas results of such experiments do show significant effects of training on metabolic responses to given absolute submaximal exercise tasks, it is probably inappropriate to make conclusions about substrate utilization in marathon running from studies using ergometry at given submaximal power outputs.

Though apparently surprising, our results indicating that experienced marathoners combusted predominantly carbohydrates corroborate those of others (Table 2). In their reports Bosch et al. (4), Scrimgeour et al. (36), and Costill (11) have consistently observed high ($\geq$0.90) gas exchange R values on athletes during long-distance running. Thus, our results are actually consistent with those of previous reports that are, in the aggregate, contradictory to the concept that marathon running is dependent upon lipid oxidation. If the simple average of respiratory gas exchange R in all reports (Table 2) is 0.91, then approximately 71% of the energy for a marathon is from carbohydrate, and 29% from lipid.

Probably the strongest support for a training-induced enhancement of fat utilization comes from observations of a lower R across trained muscle and a diminished net glycogen depletion in trained active muscle during a given power output (17). However, competitive athletes work at higher absolute and relative power outputs

than prior to training (40). We believe it is the case that athletes train and compete at high exercise intensities and remain dependent on carbohydrate as opposed to lipid oxidation. The biochemical and metabolic adaptations described previously (16,17,21) are likely important when they can affect a degree of glycogen sparing during activities such as marathon running. Additionally, training-induced adaptations in the ability to use fat as a fuel are important because they may allow some measure of exercise performance in the face of muscle glycogen depletion, and because energy can be provided in the post-exercise, glycogen depleted state.

Total Carbohydrate Combustion During a Marathon

In the present study blood glucose concentrations were maintained despite the fact that in the fast group most (60–100%) of the total energy expenditure was from carbohydrates. With an active muscle glycogen content of approximately 375 g (1500 kcal) (22), and a fasted liver content of 100 g of glycogen (400 kcal) (33), it would not be possible to sustain exercise at a $\dot{V}O_2$ between 2.5 and 3.0 $l \cdot min^{-1}$ and gas exchange R of greater than 0.90 for 2 hr 45 min, unless glycogen reserves in inactive muscle and other tissues were mobilized. Though not extensively investigated, glycogenolysis with elevated net lactate release in inactive muscle beds has been demonstrated during exercise and recovery (2).

Our study is similar to other reports on the metabolic response to marathon running in that we failed to measure muscle masses and glycogen levels in active and inactive muscle beds. For this reason, we were required to rely on previously measured values of muscle (22) and liver (33) glycogen levels to estimate available carbohydrate reserves. The value we selected for glycogen content of active muscle ($25 g \cdot kg^{-1}$) represents a value higher than normal and assumes effects of training and high carbohydrate diet. The value we selected for liver glycogen content (100 g) is probably far in excess of that actually present in the overnight-fasted subjects we studied. Hepatic glycogen stores are generally considered to be depleted in the morning after an overnight fast (35).

In our study, subjects did not "carbohydrate load", but rested and ate their normal diet the day prior to the treadmill marathon. Thus, we have reason to believe that the assumed values we used for muscle glycogen concentration in active muscle prior to the marathon were appropriate, if not inflated. However, if active muscle mass was a third (5 kg) greater than estimated, then our conclusion about global mobilization of body glycogen reserves during marathon running would have to be modified if glycogen content did reach $25 g \cdot kg^{-1}$. However, our conclusion about CHO dependence during marathon running would not be affected.

Summary

In summary, the present investigation provides data which indicate that experienced marathoners rely predominantly on carbohydrate fuel sources. Further, a relationship appears to exist between speed of running and rate of CHO oxidation. Though perhaps surprising, our results are largely in agreement with literature values. Additionally, the amount of carbohydrate combusted may exceed that in the liver and active muscle beds. Therefore, the sources of carbohydrate oxidation in active muscles during the marathon is unaccounted for and leaves an opportunity for the role of the lactate shuttle (6) in marathon running. The level of effort required to complete a marathon may require utilization of glycogen, lipids, and amino acids in diverse pools.

Research supported by the University of California Fitness Evaluation Program. The authors thank G. Reaven for assistance with insulin assays and M. Huie and J. Mercier for critical commentary in review of our manuscript.

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