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REGULATION OF CARBOHYDRATE AND FAT METABOLISM DURING AND AFTER EXERCISE

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ABSTRACT

During vigorous exercise, carbohydrate, in the form of muscle glycogen and blood glucose, is the primary energy source, whereas fatty acids play a secondary, carbohydrate-sparing role. In the postabsorptive state, nearly all of the carbohydrate used during exercise comes from muscle and liver glycogen. The size of these glycogen stores plays a major role in determining how long vigorous endurance exercise can be performed if other causes of fatigue, such as dehydration or heat exhaustion, are avoided. In a "fight or flight" situation, individuals whose muscles are glycogen depleted are helpless, as they are unable to either run or fight. The same is true of someone who has become markedly hypoglycemic as a result of liver glycogen depletion.

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During prolonged exercise, if sufficient carbohydrate is ingested hypoglycemia does not develop (12). Other than that, the major metabolic consequence of carbohydrate ingestion during exercise is an increased utilization of carbohydrate and a decreased utilization of fatty acids for the oxidative generation of energy. In the postabsorptive state, i.e. when there is no carbohydrate in the intestinal tract, plasma glucose is maintained by glycogenolysis of liver glycogen stores and by hepatic gluconeogenesis. The major gluconeogenic substrates during exercise are lactate and alanine, most of which come from breakdown of skeletal muscle glycogen, and from glycerol provided by hydrolysis of triglycerides. Provision of glucose via gluconeogenesis delays the development of hypoglycemia but does not occur rapidly enough to prevent it when hepatic glycogen stores are depleted during prolonged exercise. During exercise of an intensity that can be maintained for an hour or longer, the blood glucose level stays remarkably constant, until liver glycogen stores are depleted and hypoglycemia starts to develop (13, 24, 25, 87). Afferent neural feedback from the working muscles probably plays a major role in regulating the increase in glucose production during exercise (56). Increases in glucagon and catecholamine secretion and a decrease in insulin concentration are responsible for protection against hypoglycemia during prolonged exercise, with the changes in glucagon and insulin playing a primary role, and with catecholamines playing a secondary role (41, 60, 84, 85, 87). Although it is well documented that these factors are involved, how they are precisely regulated so as to result in an increase in the rate of glucose production that precisely matches the rate of glucose utilization remains a mystery.

Regulation of Plasma Free Fatty Acids During Exercise

In contrast to the precise regulation of hepatic glucose production and plasma glucose concentration during exercise, lipolysis of triglycerides in the adipocytes, the release of free fatty acids (FFA) into the bloodstream, and plasma FFA concentration are not closely matched to energy needs. Plasma FFA generally fall initially during vigorous exercise and then during prolonged exercise rise progressively as the result of increases in the rates of lipolysis in, and release of FFA from, adipocytes. The increases in lipolytic rate and FFA release are mediated by both the rise in catecholamine levels and the decrease in plasma insulin concentration that occur during exercise and can result in increases in plasma FFA to levels as high as ~1.5 mM after 2 h or more of exercise. Because the rate of FFA uptake and oxidation by muscle is a function of plasma FFA concentration, the contribution of plasma FFA

oxidation to the working muscles' energy needs progressively increases. Surprisingly, during 2 h of exercise requiring ~50–75% of maximal oxygen uptake capacity ($V0_{2max}$), plasma FFA generally provide less than one half of the fat oxidized, with the remainder coming from intramuscular triglyceride stores (48, 53, 61, 79). This finding is counterintuitive, as muscle triglyceride stores are small, whereas adipose tissue triglyceride stores contain many times more energy than can be utilized during a bout of exercise, even in a very lean individual. The contribution of plasma fatty acids increases during prolonged exercise as the result of the progressive increase in plasma FFA, whereas the contribution of muscle triglycerides decreases as they are gradually depleted (79).

Exercise Intensity and Utilization of Plasma Glucose and FFA

Terms such as moderate-intensity, high-intensity, and vigorous exercise must be defined relative to the individual's maximal capacity for aerobic exercise if they are to have relevance to the metabolic response. A given absolute exercise intensity that is very light for one individual can be impossibly hard for another. $V0_{2max}$ is a measure of the highest rate of ATP generation via oxidative phosphorylation that an individual can attain during exercise of progressively increasing intensity. $V0_{2max}$ differs greatly between individuals, from values as low as 12 $ml \bullet kg^{-1} \bullet min^{-1}$ in some sedentary old people to as high as 84 $ml \bullet kg^{-1} \bullet min^{-1}$ in some highly trained young male athletes. Factors that determine the differences in $V0_{2max}$ among healthy people include level of exercise training, heredity, age, and sex. The intensity of exercise can also be defined in terms of the lactate threshold (LT), which is the percentage of $V0_{2max}$ at which a significant increase in blood lactate concentration occurs (31, 51).

Large differences exist between individuals in their metabolic responses to exercise. In healthy young people these responses are determined largely by genetic factors and level of physical training. The higher the level of training, the greater the proportion of energy derived from fat oxidation at any submaximal (i.e. requiring less than $V0_{2max}$) exercise intensity. Generally, oxidation of plasma FFA can provide most of the energy needed during mild exercise (i.e. requiring less than ~30% of $V0_{2max}$), with little or no net utilization of intramuscular substrates and with minimal oxidation of plasma glucose (Figure 1) (57, 79). During moderately intense exercise, requiring ~50–70% of $V0_{2max}$ and lasting ~60–120 min, fat oxidation provides roughly one half (in the 40–60% range) of energy (13, 53, 61, 79), with intramuscular triglycerides providing 50% or more of the total FFA oxidized (Figure 1) (53, 61, 79). Release of FFA from adipose tissue occurs at a similar or slightly slower rate at 65 compared with 25% of $V0_{2max}$ (79). During high-intensity aerobic exer-

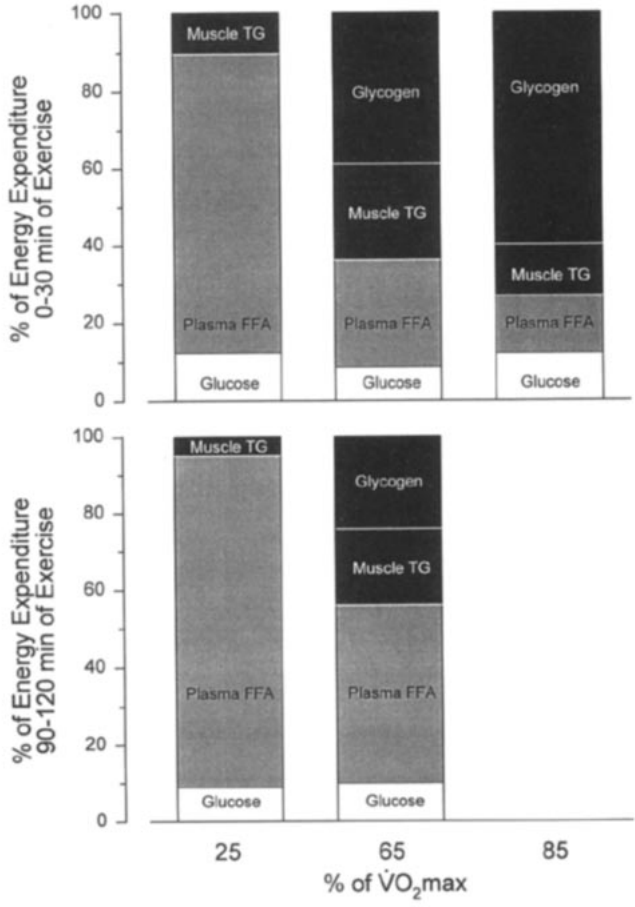

Figure 1 Effects of exercise intensity and duration on substrate utilization. Relative contributions of plasma glucose, plasma free fatty acids (FFA), muscle triglycerides (TG), and muscle glycogen to energy expended during 0–30 min of exercise performed at 25, 65, and 85% of maximal aerobic power ($V0_{2max}$) and during 90–120 min of exercise performed at 25 and 65% of $V0_{2max}$. (Adapted from Reference 79.)

cise, requiring ~75–85% of $V0_{2max}$ (above the LT), release of FFA from adipose tissue into plasma is lower than during mildly or moderately intense exercise, and both muscle triglyceride and plasma FFA oxidation are lower than at ~50–70% of $V0_{2max}$ (below the LT) (53, 79). One explanation proposed for the decrease in FFA release from adipose tissue when exercise intensity increases above the LT is that lactate increases FFA resterification (6, 49). It is also possible that vasoconstriction in adipose tissue may result in decreased

transport of FFA out of the adipose tissue (79, 80). Another mechanism, in addition to the decreased availability of plasma FFA, that may play a role in the lower rate of fat oxidation during high- as compared with moderate-intensity exercise is an increased competition of pyruvate-derived acetyl-coenzyme (Co) A with fatty acid–derived acetyl-CoA for entry into the citrate cycle.

During moderately intense exercise (~50–70% of $V0_{2max}$; below LT), plasma glucose provides 15–35% of the carbohydrate oxidized, and muscle glycogen provides the rest (Figure 1). Although carbohydrate and fat oxidation contribute roughly equally to the energy expended during moderate-intensity exercise, carbohydrate is the predominant substrate during high-intensity exercise, above the LT, providing about two thirds of the total energy (Figure 1) (53, 79). Plasma glucose and muscle glycogen utilization increase in response to high-intensity exercise, with plasma glucose making up roughly one fifth of the carbohydrate oxidized. At any absolute submaximal exercise intensity, the proportion of total energy derived from fat oxidation is greater in the trained (i.e. athletically conditioned) state than in the untrained state, resulting in slower depletion of glycogen stores in the trained state. During moderately intense exercise that can be performed for a long time, i.e. ~50–70% of $V0_{2max}$ and below LT, the contributions of intramuscular glycogen and triglycerides to the energy used by the working muscles progressively decrease, whereas the utilization of plasma substrate, particularly FFA, progressively increases (Figure 1) (79).

Although the relative exercise intensity expressed as a percentage of $V0_{2max}$ is of primary importance in regulating carbohydrate utilization, the absolute work rate is also important, as it determines the total energy requirement. For example, an individual with a $V0_{2max}$ of 60 $ml \bullet kg^{-1} \bullet min^{-1}$ has to generate ATP via aerobic metabolism at a rate twice as fast as an individual with a $V0_{2max}$ of 30 $ml \bullet kg^{-1} \bullet min^{-1}$ when both are working at the same percentage of $V0_{2max}$. Assuming they are using the same fuel mix, the individual with a $V0_{2max}$ of 60 $ml \bullet kg^{-1} \bullet min^{-1}$ would utilize twice as much glycogen per hour as the one with a $V0_{2max}$ of 30 $ml \bullet kg^{-1} \bullet min^{-1}$.

Effect of Nutritional State on Utilization of Plasma Glucose and FFA

A major factor that determines what the relative rates of plasma glucose and FFA utilization are and how long a given vigorous submaximal level of exercise can be maintained is nutritional status. Fasting or eating a low-carbohydrate diet results in a reduced capacity for vigorous prolonged exercise (10, 11, 68). A major reason for this decrease in endurance is that hypoglycemia develops rapidly because of a low initial hepatic glycogen concentration, and because gluconeogenesis cannot keep pace with glucose utilization. Hypogly-

cemia would develop even more rapidly if the rate of glucose utilization were not reduced as a result of increased plasma FFA levels and more rapid fat oxidation. There is evidence that the ability of individuals on a low-carbohydrate diet to perform prolonged low to moderate–intensity exercise is improved somewhat if they are allowed to adapt to the diet for a few weeks (65, 66). In individuals eating a high-carbohydrate diet, glucose utilization is increased during exercise in the fed state; despite the increase in glucose utilization, hypoglycemia is delayed as the result of a high initial hepatic glycogen concentration (10, 11, 68).

Carbohydrate ingestion during prolonged exercise prevents the development of hypoglycemia and can delay the development of fatigue (12). However, carbohydrate ingestion during prolonged moderately intense exercise does not spare muscle glycogen (23, 37); the increased oxidation of glucose just results in a proportional decrease in fat oxidation. As mentioned earlier, vigorous exercise is no longer possible when muscle glycogen stores are depleted. How then do triathletes, long-distance bicycle racers, ultramarathoners, and various other athletes perform strenuous exercise for many hours longer than could be supported by their initial glycogen stores? We think that such prolonged, strenuous exercise without glycogen depletion is made possible by combining carbohydrate ingestion with brief rest periods or with periods during which the exercise intensity is decreased sufficiently so that rapid net glycogen synthesis can occur before strenuous glycogen-depleting exercise is resumed (21, 59). The phenomenon of glycogen resynthesis during continued lower-intensity exercise is discussed further below, in sections dealing with muscle glycogen metabolism.

During exercise, elevation of plasma FFA has a powerful glycogen-sparing effect (22, 40, 75), which is probably mediated via the glucose–fatty acid cycle (69, 70, 74). This effect has been demonstrated by raising plasma FFA by means of an all-fat meal followed by injection of heparin (22, 40, 75). However, the only way plasma FFA can be increased by dietary means is with a low-carbohydrate diet or by fasting, both of which are counterproductive because they result in glycogen depletion.

Effects of Endurance Exercise Training on Utilization of Plasma Glucose and FFA

Endurance exercise training results in a sparing of plasma glucose and liver glycogen (2, 13, 34). Depletion of glycogen stores results in the inability to continue to exercise vigorously. Carbohydrate sparing is, therefore, one of the most important mechanisms by which training increases the ability to perform prolonged strenuous exercise. The slower utilization of glycogen is made possible by training–induced adaptations to endurance exercise that result in

an increase in the proportion of energy derived from fat oxidation during prolonged exercise. In a study in which plasma glucose turnover was measured with [^{13}C]glucose in the same individuals before and after endurance exercise training, Coggan et al (13) found that the rates of plasma glucose appearance, disposal, and oxidation were all reduced by ~33% during exercise at the same absolute intensity after training. The slower rate of glucose production by the liver in the trained state is due to reductions in the rates of both hepatic glycogenolysis and gluconeogenesis (16).

The same absolute exercise intensity represents a lower relative exercise intensity after training has resulted in an increase in $V0_{2max}$. The proportion of energy derived from carbohydrate generally increases as relative exercise intensity is raised, and carbohydrate becomes the predominant fuel during strenuous exercise (Figure 1). The decreased glucose utilization at the same absolute exercise intensity after training can therefore be partly explained on the basis of a lower relative exercise intensity and, therefore, a reduced metabolic stress. However, athletes train and compete at high relative exercise intensities. It is, therefore, of interest to know whether or not the adaptations to exercise training also result in a sparing of carbohydrate at the same relative exercise intensity. This comparison is complicated by the larger total energy requirement and by the involvement of a larger muscle mass at the same relative exercise intensity in the trained state. In a study in which endurance exercise training increased $V0_{2max}$ from 43 to 55 ml•kg^{-1}•min^{-1}, the respiratory exchange ratio during exercise requiring 75% of $V0_{2max}$ was 0.94 before training and 0.91 at the same relative (27% higher absolute) exercise intensity after training (47). Thus, carbohydrate oxidation accounted for ~79% of the energy used at 75% of $V0_{2max}$ before training, and only ~69% after training. Essentially all of the 27% increase in energy utilization was accounted for by oxidation of fat. In the same study, highly trained endurance athletes, who had a $V0_{2max}$ of 70 ml•kg^{-1}•min^{-1}, had a respiratory exchange ratio of 0.88 ± 0.01 during exercise at 75% of $V0_{2max}$, indicating that only ~58% of their energy came from carbohydrate oxidation (47).

Measurement of the respiratory exchange ratio does not, of course, provide information regarding the relative contributions of decreased blood glucose and decreased muscle glycogen utilization to the carbohydrate-sparing effect of training in this study. However, in a recent study comparing endurance-trained and untrained subjects exercising at 80% of $V0_{2max}$, Coggan et al (14) found that utilization of blood glucose was 19% lower in the trained subjects, even though they were working at a 40% higher absolute exercise intensity than were the untrained individuals. These results provide evidence that training-induced adaptations spare liver glycogen even at the same relative exercise intensity.

The mechanisms responsible for the slower rate of glucose uptake by the

muscles, and for the closely matched decrease in hepatic glucose production, at the same exercise intensity in the trained state are still unexplained. Especially surprising is the finding that glucose uptake by working muscles during exercise at the same absolute intensity is lower after training (13), even though training increases the number of glucose transporters (GLUT4) in skeletal muscle (46), with concomitant increases in the capacities for insulin-stimulated and for contraction-stimulated glucose transport (72, 78). It does seem clear, however, that the decrease in glucose uptake is not mediated by the glucose–fatty acid cycle, as citrate and glucose-6-phosphate concentrations are lower in muscle at the same exercise intensity in the trained than in the untrained state (15).

The decreased carbohydrate utilization during exercise at the same intensity after training is compensated for by an increase in fat oxidation. However, contrary to what one might expect, plasma FFA turnover and oxidation are lower after training because of a slower rate of lipolysis in adipose tissue. In a study in which 13 subjects were evaluated during 90–120 min of exercise before and after 12 weeks of training that elicited a 24% increase in $V0_{2max}$, the rate of plasma FFA oxidation was ~28% lower after training (61). The exercise required ~58% of $V0_{2max}$ before training and 46% of $V0_{2max}$ after. In untrained subjects, approximately 40% of the energy utilized during the exercise was derived from fat oxidation; in trained subjects performing the same bout of exercise, ~60% of the energy was obtained from fat oxidation. As a consequence of the decreased oxidation of plasma FFA, the contribution of plasma FFA to CO_2 production from fat oxidation declined from 43 to 23%. Thus, muscle triglycerides provided ~50% of the FFA oxidized in the untrained state and more than 75% of the FFA oxidized in the trained state (61). Both the calculated (61) and the measured (48) utilization of muscle triglycerides were increased roughly twofold after 12 weeks of training.

What is the explanation for the surprising finding that, during the first 2 h of moderately intense exercise, trained individuals preferentially utilize muscle triglycerides even though the triglyceride stores in human muscles are small, whereas large amounts of fat are stored in fat cells? We think this is a compensatory mechanism necessitated by the decreased availability of plasma FFA as the result of a reduced rate of lipolysis in the adipocytes. Exercise training rapidly induces a marked blunting of the catecholamine response to exercise of a given intensity (58, 86) and, therefore, a reduced stimulus for lipolysis of fat cell triglycerides. Apparently, lipolysis of intramuscular triglycerides is less dependent on beta-adrenergic stimulation.

MUSCLE GLYCOGEN

The development of exhaustion during prolonged strenuous exercise is delayed by raising muscle glycogen, and occurs more rapidly when muscle glycogen

concentration is low (3, 55), if other causes of fatigue such as dehydration or heat-exhaustion are avoided. Although the requirement for muscle glycogen during vigorous exercise was first described in 1967 (3), its biochemical basis, i.e. why exhaustion occurs when the other substrates (i.e. fatty acids and blood glucose) are still available, is not yet completely explained. In the case of blood glucose, the likely explanation is that its rate of entry into the working muscles is not sufficiently rapid to compensate for the glycogen deficiency. The reason for the inability of skeletal muscle to increase fatty acid oxidation sufficiently to compensate for glycogen depletion is less clear. One possibility is that citrate cycle activity decreases as a result of depletion of citrate cycle intermediates when the rate of glycolysis drops below some critical level and fatty acids become the predominant substrate for oxidation by the mitochrondria. Citrate cycle intermediates are continuously drawn off into other metabolic pathways, and the citrate cycle would run down if one or more of the cycle intermediates were not replenished at a sufficiently rapid rate.

The process of citrate cycle replenishment is called anaplerosis. The major anaplerotic precursor is pyruvate, which can be converted to oxaloacetate in the pyruvate carboxylase reaction in most mammalian cells. Skeletal muscle apparently does not contain pyruvate carboxylase, but it does contain malic enzyme (62), which, by converting pyruvate to malate, can replenish the citrate cycle. In contrast, fatty acid metabolism does not provide any anaplerotic precursors. Thus, in glycogen-depleted muscles, the rate of pyruvate production may be too low to prevent depletion of citrate cycle intermediates. This could prevent fat oxidation from occurring rapidly enough to provide the energy (ATP) required for strenuous exercise. This possibility needs further investigation.

Muscle Glycogen Supercompensation

Because of the evidence that raising muscle glycogen improves endurance for prolonged strenuous exercise, there has been much interest in the glycogen "supercompensation" phenomenon. Glycogen supercompensation is the increase in muscle glycogen concentration, to levels far above those found in well-fed sedentary individuals, that occurs in response to carbohydrate feeding after a bout of glycogen-depleting exercise. This phenomenon is limited to the muscles involved in the exercise, as was clearly shown in the original study by Bergström & Hultman (4), which first demonstrated the glycogen supercompensation phenomenon. In this study, the two investigators performed one-legged cycle ergometer exercise and used their nonexercised legs as controls. Ingestion of a high-carbohydrate diet following the exercise resulted in an increase in muscle glycogen to roughly twice the preexercise level in the exercised muscles, whereas no significant change in glycogen concentration occurred in the muscles of the nonexercised lower extremity (4).

MECHANISMS INVOLVED IN GLYCOGEN SUPERCOMPENSATION Glycogen depletion is associated with activation of glycogen synthase (5, 26). Glycogen synthase and glycogen synthase phosphatase are both normally bound to glycogen. During exercise, glycogen is broken down and these enzymes are released from the glycogen particle; this makes the inactive glycogen synthase D available for conversion to the active I form by glycogen synthase phosphatase (81). As glycogen concentration decreases, glycogen synthase I activity increases (5). Considerable interest has focused on the possibility that the activation of glycogen synthase is responsible for muscle glycogen supercompensation after exercise. However, it has become evident that the increase in glycogen synthase activity is only involved in the early rapid component of glycogen repletion and does not play a role in glycogen supercompensation. The best evidence for this is that the activation of glycogen synthase after exercise reverses when muscle glycogen returns to the level found in resting muscles in the fed sedentary state, i.e. before glycogen supercompensation begins (17). The primary step that regulates the rate and extent of muscle glycogen accumulation is the transport of glucose across the cell membrane into the cytoplasm. Of the various studies supporting this conclusion, probably the most clear-cut is that of Ren et al (71) on transgenic mice overexpressing the GLUT1 type of glucose transporter in skeletal muscle. Basal glucose transport into muscle is increased approximately sevenfold in GLUT1 transgenic mice. This is similar to the rate of glucose transport induced by a maximally effective insulin concentration in muscles of normal wild-type mice. There is an enormous accumulation of glycogen in muscles of the GLUT1 transgenic mice to values tenfold higher than those found in fed, normal, i.e. nontransgenic, mice (71). This huge increase in muscle glycogen is entirely due to the increase in glucose uptake, as it occurs despite a 50% reduction in the active I form of glycogen synthase, clearly showing that glucose transport is normally rate limiting for glycogen synthesis.

The glycogen supercompensation that occurs following glycogen-depleting exercise also results from increased glucose transport into muscle. Muscle contractions result in a powerful stimulation of glucose transport in skeletal muscle that is independent of insulin action (45, 50, 67, 82, 83). As this acute effect of exercise wears off, it is replaced by an increased sensitivity of the glucose transport process to stimulation by insulin (76). The increase in sensitivity of the glucose transport process to insulin persists until glycogen supercompensation occurs (8). As a result of this increase in insulin sensitivity, glucose transport and glycogen accumulation are increased following exercise if sufficient glucose is available, i.e. if an adequate amount of carbohydrate is ingested. Exercise physiologists and endurance athletes refer to the increase in muscle glycogen in response to a high-carbohydrate diet following glycogen depleting exercise as carbohydrate loading.

Glucose transport across the plasma membrane is mediated by a family of glucose transport proteins. The glucose transporter in skeletal muscle that mediates insulin-stimulated and exercise-stimulated glucose transport is the GLUT4 isoform. In the unstimulated state, the GLUT4 transporters are located intracellularly and are translocated (i.e. move to and are incorporated) into the plasma membrane by the actions of insulin and contractions (27, 35, 42). There is a good correlation between GLUT4 concentration and maximally stimulated glucose transport in muscle (38). Exercise training induces an increase in GLUT4 concentration in muscle that is associated with increases in the capacity for both insulin- and exercise-stimulated glucose transport (72, 78).

An adaptive increase in GLUT4 can occur very rapidly in response to an adequate adaptive stimulus. In a study by Ren et al (72), rats were exercised by means of swimming for two 3-h-long periods separated by a 45-min rest period. Sixteen hours after the exercise, forelimb muscles, used during swimming, showed a twofold increase in GLUT4 mRNA and a 50% increase in GLUT4 protein. After a second day of the same exercise protocol, muscle GLUT4 protein was increased twofold (72). Insulin-stimulated glucose transport was also increased twofold. In muscles incubated in vitro with glucose and insulin, the rate of glycogen accumulation was twice as great in muscles of the two-day swimmers as in muscles from sedentary rats (72). It seems probable that the physiologic role of the increase in GLUT4 is to increase the rate of muscle glycogen accumulation after glycogen-depleting exercise. In an evolutionary context, the rapid increase in GLUT4 could provide a survival advantage to an animal faced with the necessity for a sustained increase in physical activity (for example invasion of its territory by predators) by making possible more rapid muscle glycogen repletion when carbohydrate is eaten during brief rest periods, or even during continued lower-intensity exercise (21). In an athletic context, this adaptation can enhance muscle glycogen repletion between strenuous prolonged daily training sessions, or during prolonged competitive events such as cross-country cycling.

Sparing of Muscle Glycogen

Because muscle glycogen depletion results in the inability to exercise vigorously, endurance can be increased not only by raising muscle glycogen concentration prior to exercise, but also by slowing the rate of glycogen depletion. This can be accomplished by increasing the proportion of energy generated by oxidation of fatty acids. The contribution of fatty acid oxidation to the energy required by the working muscles can be increased, and the rate of glycogen utilization can be proportionally decreased, either by increasing the concentration of FFA to which the muscles are exposed or by means of endurance exercise training. The glycogen-sparing effect of raising plasma

FFA has already been discussed. It has been demonstrated experimentally in rat hindlimb muscles stimulated to contract while being perfused with a high concentration of oleate (74), and during treadmill running in rats (40, 75) and humans (22) whose plasma fatty acids were raised with a fat meal followed by injection of heparin. However, these are artificial, experimental conditions; there is no physiologic way to raise plasma FFA without also causing glycogen depletion.

Endurance exercise–trained humans and experiment animals utilize less glycogen and more fat during submaximal exercise than do otherwise comparable untrained individuals (39, 44, 48, 52, 61), and this glycogen sparing plays a major role in their increased endurance. Endurance exercise training induces an increase in muscle mitochondria, which can be twofold or greater (43). As a consequence of this increase in mitochondria, there is less disturbance of homeostasis, with smaller decreases in creatine phosphate and ATP and smaller increases in adenosine diphosphate (ADP) and inorganic phosphate (P_i) at the same work rate in trained than in untrained muscle (20, 29).

REGULATION OF GLYCOGENOLYSIS Evidence from a number of studies indicates that availability of P_i limits glycogenolysis in resting muscle, and also in contracting muscle during mild and moderate work intensities (9, 32, 36, 73). Glycogenolysis is catalyzed by the enzyme glycogen phosphorylase, which mediates the phosphorolysis of the α-1,4 bond linking glucose residues in glycogen to form glucose-1-phosphate. Phosphorylase occurs in two molecular forms, phosphorylase *b*, which is inactive in muscle under most physiologic conditions, and phosphorylase *a*, which is the active form. Phosphorylase *b* is converted to the *a* form by phosphorylase kinase, which requires Ca^{2+} for catalytic activity (7, 33). Phosphorylase kinase exists in a dephosphorylated, less-active *b* form that is converted to the more active *a* form by protein kinase A. Protein kinase A is activated by increases in cyclic AMP in response to catecholamines (33). Phosphorylase kinase *a* can activate phosphorylase at the low Ca^{2+} concentration found in the cytosol of resting muscle, whereas phosphorylase kinase *b* is inactive in resting muscle but becomes active at the Ca^{2+} concentration attained in contracting muscle (7, 28, 33). The Ca^{2+} released from the sarcoplasmic reticulum at the onset of muscle contractile activity activates phosphorylase kinase *b*, which in turn causes a large increase in the proportion of phosphorylase in the active *a* form and a burst of glycogenolysis (7). The activation of phosphorylase by the Ca^{2+} mechanism at the onset of exercise results in an overshoot of glycogen breakdown, in excess of the muscles' energy requirement, resulting in accumulation of lactate (20, 32, 73). After a few minutes, the activation of phosphorylase reverses despite continued contractile activity, with a return of the proportion of phosphorylase that is in the *a* form to, or below, that found in resting muscle, i.e. ~8–12% phospho-

rylase *a* (1, 9, 18, 77). The mechanisms that mediate the reversal of phosphorylase activation despite continued contractile activity are not well understood. One process that may be involved is release of the phosphorylase bound to the glycogen particle-enzyme-sarcoplasmic reticulum complex (30, 63). This complex also contains phosphorylase kinase, and it has been hypothesized that phosphorylase is released during glycogenolysis, resulting in uncoupling from phosphorylase kinase and the Ca^{2+}-activating mechanism (30). However, other factors that have not been elucidated also appear to be involved (19).

Despite the rapid reversal of phosphorylase activation, prolonged strenuous exercise can result in a progressive decrease in muscle glycogen stores until, at the point of exhaustion, they are almost completely depleted (3). As mentioned earlier, roughly 10% of the phosphorylase in resting muscle is in the active *a* form, and a similar level of phosphorylase *a* activity is present in contracting muscle after the acute activation of phosphorylase has reversed (9, 18). Although the phosphorylase *a* activity present in resting muscle is sufficient to mediate moderately rapid glycogenolysis, the low concentration of P_i in the cytoplasm prevents net glycogenolysis in well-oxygenated resting muscle. Raising the concentration of P_i in the cytoplasm by making muscle hypoxic causes sustained glycogenolysis without any increase in phosphorylase *a* above the basal level of ~10% (73). Similarly, in exercising muscles in which phosphorylase activation has reversed and the percentage of phosphorylase *a* has returned to the basal level, the increase in P_i concentration caused by muscle contractions makes possible continued glycogenolysis at a slower rate. The finding that cytosolic P_i concentration limits the rate of glycogenolysis explains how the glycogen-sparing effect of exercise training is mediated.

Glycogen sparing in the endurance exercise–trained state is manifested both as a smaller initial burst of glycogenolysis and as a slower rate of glycogen depletion during prolonged exercise (20, 32, 34, 54). The initial, rapid phase of glycogenolysis is smaller in endurance-trained than untrained skeletal muscles, despite no difference in the extent of phosphorylase activation as reflected in the increase in the percentage of phosphorylase *a* (32). However, as a result of the adaptive increase in mitochondria in trained muscles (43), the increase in P_i concentration in response to the same contractile activity is smaller (20, 29, 32). The steady state concentration of P_i maintained in the muscles during continued contractile activity is also lower in trained muscles. The lower level of P_i in trained muscles provides an explanation for both the smaller initial burst of glycogenolysis and the slower rate of glycogen depletion during continued contractile activity (20, 32). As a result of the slower rates of glycogenolysis and glycolysis in the trained state, the rate of acetyl-CoA formation from carbohydrate is reduced, and fatty acid oxidation increases to compensate for the reduced availability of carbohydrate for oxidative metabolism.

The mechanisms responsible for mediating the increase in the contribution of fat oxidation to energy metabolism in the trained state have not been fully elucidated. One factor that probably plays a role in making possible the increase in fat oxidation is the adaptive increase in the enzymes involved in fatty acid transport and oxidation in skeletal muscle in response to exercise training (44, 64). The factors responsible for the proportional increase in fat oxidation to compensate for the decrease in carbohydrate, i.e. pyruvate availability, likely include (*a*) reduced competition for the available CoA by the pyruvate dehydrogenase reaction, and (*b*) reduced competition of pyruvate-derived acetyl-CoA with fatty acid–derived acetyl-CoA for reaction with oxalocetate in the citrate synthase reaction.

CONCLUSION

Our understanding of the regulation of carbohydrate and fat metabolism during and after exercise has increased considerably in recent years. Much of this progress has been made possible by the use of stable isotope tracers to study turnover of glucose and fatty acids in humans during exercise. In addition, major insights regarding how glycogenolysis in muscle is regulated, and the mechanism by which the rate of glycogenolysis is geared to exercise intensity, have come from in situ and in vitro studies on rat skeletal muscles. Despite these recent advances, a number of fundamental questions remain to be answered. These include: Why is muscle glycogen necessary for exercise of moderate and high intensities? How is hepatic glucose production so accurately geared to muscle glucose uptake during prolonged mild to moderate–intensity exercise? What are the mechanisms responsible for the lower rate of glucose uptake by the exercising muscles in the trained as compared with the untrained state? How is the rate of fatty acid oxidation increased to compensate for the slower rate of carbohydrate utilization during submaximal exercise after adaptation to endurance exercise training? These are difficult questions, and it will be interesting to follow the progress that is made toward answering them.

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