Exercise Metabolism in Health and Disease

5

Anastassios Philippou, Costas Chryssanthopoulos, Maria Maridaki, George Dimitriadis, and Michael Koutsilieris

Abbreviations

1-RM	One-repetition maximum
Acetyl-CoA	Acetyl coenzyme A
ADP	Adenosine diphosphate
AMP	Adenosine monophosphate
AMPK	AMP-activated protein kinase
AT	Anaerobic threshold
ATP	Adenosine triphosphate
ATPase	Adenosine triphosphatase
BCAA	Branched-chain amino acids
BCOADH	Branched-chain 2-oxoacid dehydrogenase
CK	Creatine kinase
CPK	Creatine phosphokinase
Cr	Creatine
$FADH_2$	Flavin adenine dinucleotide
FAT/CD 36	Fatty acid translocase
FATP	Fatty acid transport protein
FFA	Free fatty acids
FT	Free testosterone
G-6-P	Glucose-6-phosphate
GH	Growth hormone
GLUT4	Glucose transporter type 4
GSDV	Glycogen storage disease type V
HbA1c	(Glycated) hemoglobin A1c
HDL-C	High-density lipoprotein cholesterol
HGP	Hepatic glucose output
HIF-1	Hypoxia-inducible factor-1
HIIT	High-intensity interval training
HSL	Hormone-sensitive lipase
IGF-1	Insulin-like growth factor-1
IL	Interleukin
IMP	Inosine monophosphate
LDH	Lactate dehydrogenase
LDL-C	Low-density lipoprotein cholesterol
LDM	Low-density microsomes
LT	Lactate threshold
MCT1	Monocarboxylate transporter 1
mPTP	Mitochondrial permeability transition pore
MTGL	Muscle triacylglycerol lipase
NAD	Nicotinamide adenine dinucleotide
NADH	Reduced form of NAD
NH_4^+	Ammonium
NO	Nitric oxide

A. Philippou · C. Chryssanthopoulos
M. Koutsilieris (✉)
Department of Physiology, Medical School, National and Kapodistrian University of Athens, Athens, Greece
e-mail: mkoutsil@med.uoa.gr

M. Maridaki
Department of Sports Medicine & Biology of Physical Activity, Faculty of Physical Education and Sport Science, National and Kapodistrian University of Athens, Athens, Greece

G. Dimitriadis
Second Department of Internal Medicine-Research Institute and Diabetes Center, "Attikon" University Hospital, Medical School, National and Kapodistrian University of Athens, Athens, Greece

P. Kokkinos, P. Narayan (eds.), *Cardiorespiratory Fitness in Cardiometabolic Diseases*,
https://doi.org/10.1007/978-3-030-04816-7_5

PCr	Phosphocreatine
PDH	Pyruvate dehydrogenase complex
PFK	Phosphofructokinase
PGC-1α	Peroxisome proliferator-activated receptor gamma coactivator 1-alpha
PHOS	Glycogen phosphorylase
RER	Respiratory exchange ratio
ROS	Reactive oxygen species
SHBG	Sex hormone-binding globulin
SIRTs	Sirtuins
SNARE	Soluble *N*-ethylmaleimide-sensitive factor attachment protein receptors
T1D	Diabetes mellitus type 1
T2D	Diabetes mellitus type 2
TCA	Tricarboxylic acid cycle
TGs	Triglycerides
VCO_2	Volume of carbon dioxide expired
V_E	Volume of air inspired or expired
VLDL-C	Very low-density lipoprotein cholesterol
VO_2	Volume of oxygen uptake
VO_2max	Maximal oxygen uptake

Exercise Metabolism: An Overview

In vertebrates, movement is accomplished by the contraction of skeletal muscles attached to bones via tendons. The compound adenosine triphosphate (ATP), considered the energy currency of the human body, provides the energy requirements of the working muscles. The chemical energy incorporated into the ATP is released as ATP is hydrolyzed by adenosine triphosphatase (ATPase), providing the energy for the thin myofilaments of actin to slide on the thick myofilaments of myosin. During the initial 7–10 s of maximum or near maximum physical effort, the ATP requirements are met almost exclusively by the energy compound phosphocreatine (PCr) stored within the muscles. As ATP is degraded to adenosine diphosphate (ADP), the phosphate from the stored PCr binds to ADP and ATP is formed. As activity continues beyond approximately 10 s, the limited supplies of PCr are exhausted, and the intensity of the activity (or exercise) begins to decline. However, this allows the necessary time for the glycolytic pathways to maximize their capacity to form ATP and become the predominant supplier of energy for the working muscles for the next few minutes.

Anaerobic metabolism occurs in the cytosol. The formation of ATP via the glycolytic pathways (anaerobically) involves the degradation of glycogen and glucose to pyruvate and lactate (Fig. 5.1). Muscle glucose and glycogen primarily serve the energy needs of the host muscle. Glucose entering the myocytes is phosphorylated by the enzyme hexokinase to form glucose-6-phosphate (G-6-P). This is an irreversible step as the muscle lacks the enzyme glucokinase (found in the liver) responsible for dephosphorylation of G-6-P and the exit of glucose from the cell into the bloodstream. Thus, muscle glycogen can be degraded to glucose as needed by the host muscle but cannot exit the muscle cells (enter the bloodstream) to serve the energy needs of other organs.

Aerobic metabolism takes place in the mitochondrion. ATP is formed either from pyruvate entering the mitochondrion or acetyl coenzyme A (Acetyl-CoA) formed from blood-borne or intramuscular fatty acids through β-oxidation (Fig. 5.1). Aerobic metabolism is a much more efficient process, and the amount of ATP formed is much higher than the ATP formed anaerobically.

Proteins in the form of amino acids may also support ATP resynthesis via aerobic metabolism. However, the contribution of amino acid oxidation to total energy demand is almost negligible during high-intensity exercise, whereas during prolonged exercise, it accounts for about 3–6% of the total ATP resynthesis [1, 2]. Nevertheless, amino acid oxidation may contribute more to total energy expenditure especially when body carbohydrate levels are low [3].

ATP concentrations are maintained fairly constant, even during maximal exercise intensity as energy substrates such as PCr and muscle glyco-

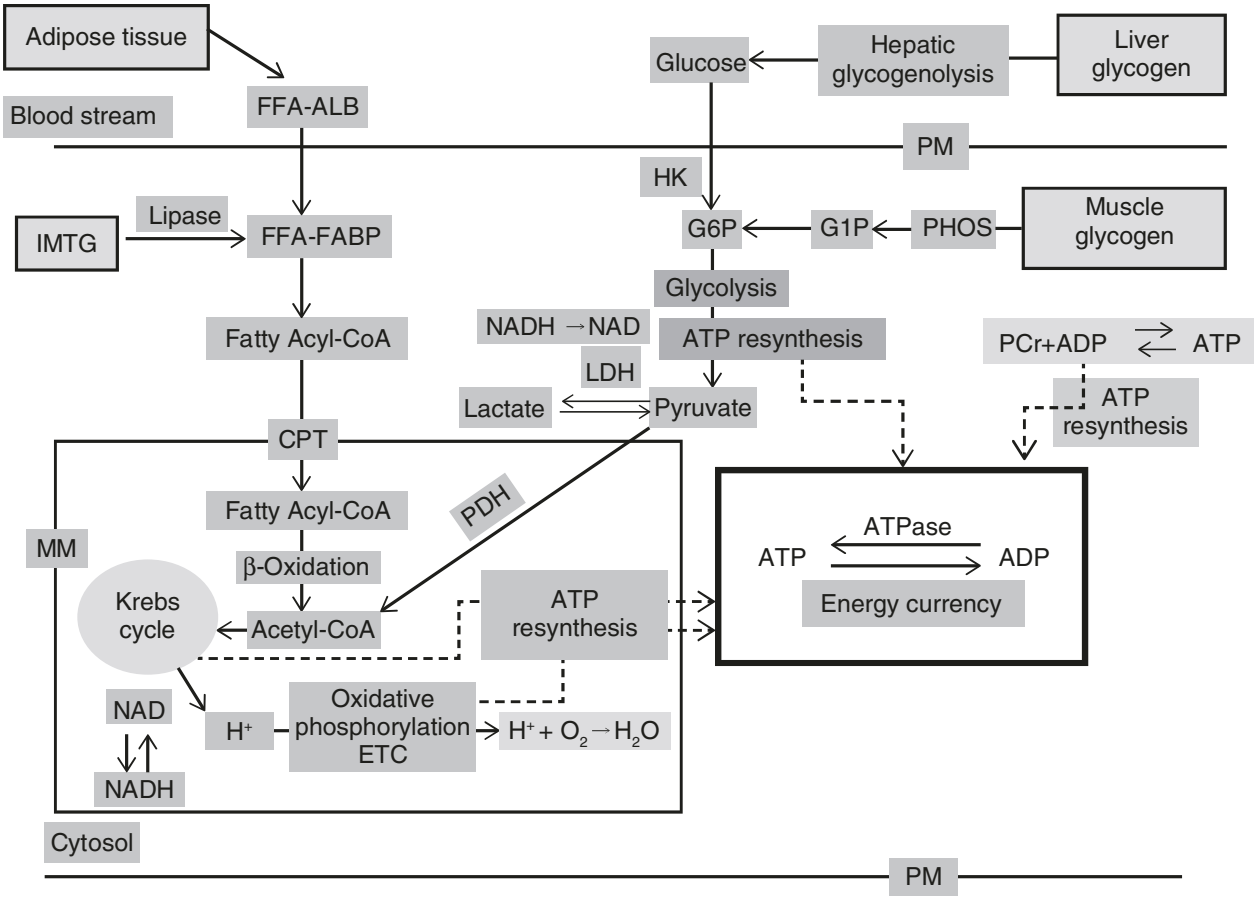


Fig. 5.1 A simplified overview of energy production in skeletal muscle. Acetyl-CoA acetyl-coenzyme A, acyl-CoA acyl-coenzyme A, ADT adenosine diphosphate, ATP adenosine triphosphate, ATPase adenosine triphosphatase, CPT carnitine palmitoyltransferase, ETC electron transport chain, FFA-ALB free fatty acids-albumin, FFA-FABP free fatty acid-fatty acid binding protein, G1P glucose-1phosphate, G6P glucose-6-phosphate, HK hexokinase, IMTG intramuscular triglycerides, LDH lactate dehydrogenase, MM mitochondrial membrane, NAD nicotinamide adenine dinucleotide, NADH nicotinamide adenine dinucleotide reduced form, PCr phosphocreatine, PDH pyruvate dehydrogenase, PHOS phosphorylase, PM plasma membrane

gen replenish the ATP utilized for the task at hand. Substrate use for ATP formation is modulated by the exercise intensity. At very high exercise intensities, PCr contribution to ATP regeneration is high. During a 30-s all-out effort of cycling or running, postexercise PCr were reduced by about 75–80%, and ATP levels by less than 30% [4, 5]. In endurance type of exercise to volitional fatigue, muscle glycogen was reduced by more than 80%, whereas ATP by only 6% [6]. Muscle fiber heterogeneity also plays a considerable role in substrate use for ATP formation and utilization [7].

The aforementioned metabolic pathways do not function independently, but in an integrative manner, where the main factor determining the relative contribution of aerobic and anaerobic metabolism is exercise intensity. If we consider a person starting exercise at a low intensity equivalent to 5 $Km.h^{-1}$, and this intensity requires a volume of oxygen uptake (VO_2) of 14 $ml.kg^{-1}.min^{-1}$, this oxygen demand is about fourfold higher than the resting of VO_2 that is about 3.5 $ml.kg^{-1}.min^{-1}$. The amount of energy for this initial stage of exercise is also supported by anaerobic metabolism, since aerobic metabolism is slow and cannot meet instantaneously the VO_2 required (Fig. 5.2). The amount of oxygen not provided, that is, illustrated in Fig. 5.2 above the VO_2 line, is referred to as oxygen deficit. If the individual continues exercise to volitional fatigue in a graded exercise intensity fashion, where intensity is increased every 3 min, a maximal oxygen uptake (VO_2max) level will be reached (Fig. 5.3).

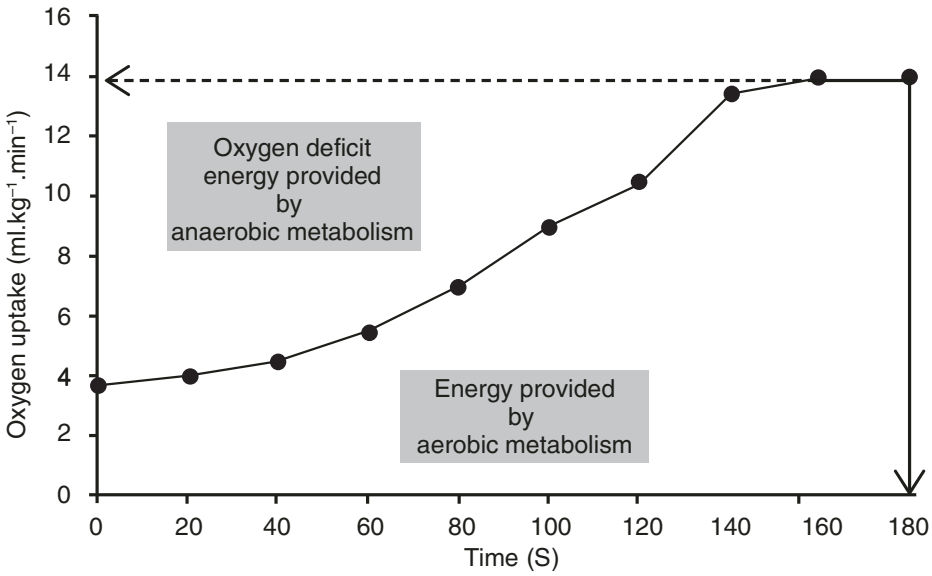
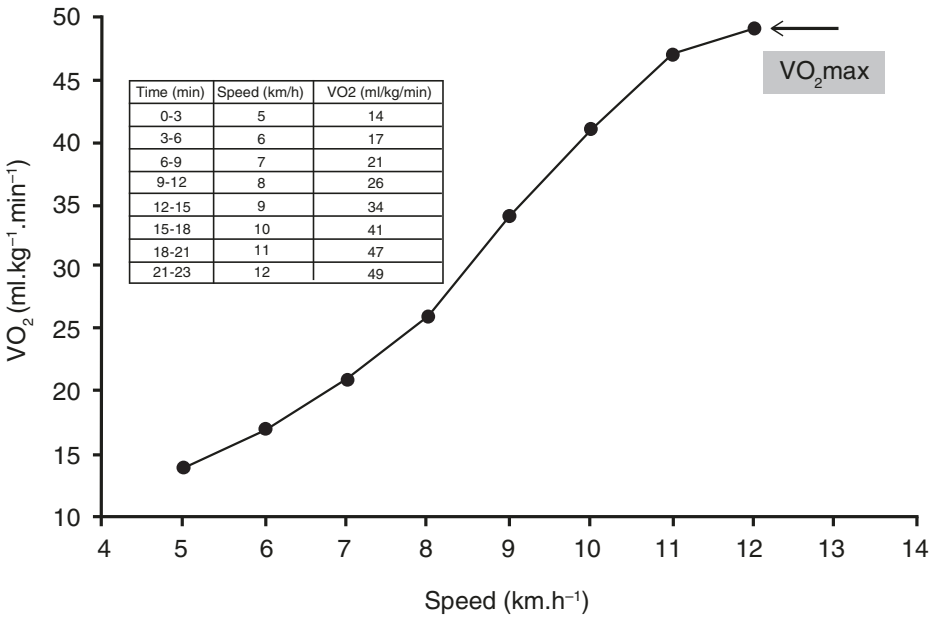


Fig. 5.2 Initial stage of a hypothetical grated exercise test of a healthy individual on a treadmill



Time (min)	Speed (km/h)	VO2 (ml/kg/min)
0-3	5	14
3-6	6	17
6-9	7	21
9-12	8	26
12-15	9	34
15-18	10	41
18-21	11	47
21-23	12	49

Fig. 5.3 Hypothetical grated exercise test to volitional fatigue of a healthy individual on a treadmill

The energy demand exercising above the VO_2max level is mainly supported by anaerobic metabolism (Fig. 5.4).

Muscle Fiber Types

Skeletal muscles are comprised by different fiber types that possess distinct morphological, histochemical, biochemical, or physiological characteristics [8]. In fact, based on myosin heavy chain gene expression, muscle fibers have an almost continuous spectrum of ATP usage and muscle contraction speeds [9, 10].

In humans, skeletal muscle fibers are broadly classified as type I (slow twitch) and type II (fast twitch). Type II fibers are further classified into three major subtypes (types IIa, IIb, and IIX) [11]. Type I fibers are, aerobically oriented fibers, designed for long-duration exercise. They have an extended capillary network and numerous mitochondria and produce a low level of force. On the other spectrum, type IIb and IIX fibers are fast twitch, have a larger diameter than type I, and therefore can produce more force. They are designed for higher exercise intensities, with their energy demands met predominantly by the glycolytic pathways, but they fatigue fast. Finally,

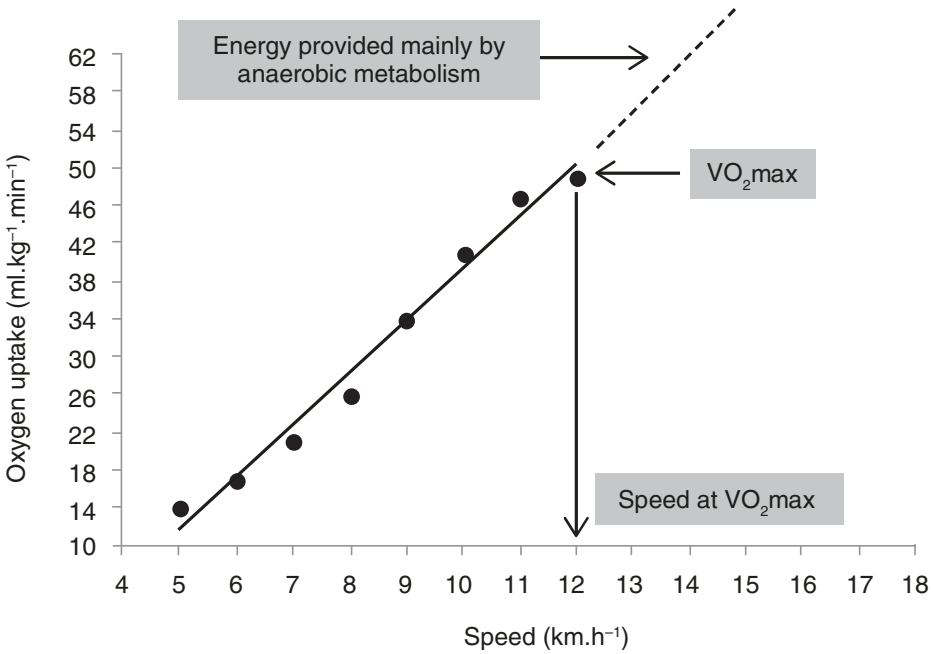


Fig. 5.4 When exercise intensity exceeds that at which VO_2max is achieved, the energy needs are met mainly by anaerobic metabolism

type IIa fibers are considered intermediate fibers, having characteristics of both type I and II [12]. The recruitment of the different fiber types is mainly dictated by the exercise intensity and the level of force developed by the muscle. For exercise intensities up to 40% of VO_2max, type I fibers are the predominant fibers recruited. As the intensity increases, progressively more type IIa fibers are recruited. For intensities >75% VO_2max, type IIa and especially IIb fibers are recruited, as intensities approach > 90% VO_2max [13]. Whether fibers can be altered as a result of chronic and specific exercise training has been scrutinized for years. The consensus is that fibers are likely to be altered to accommodate the demand imposed by the type of work. Thus, the glycolytic capacity of aerobically oriented fibers (type I) can be enhanced if these fibers are exposed to anaerobic work and vice versa [14].

In medicine, fiber typing may be important for certain fibers are prone to disease genetic myopathies, while others seem to be resistant. Some of these diseases include Duchenne muscle dystrophy, myotonic dystrophy, facioscapulohumeral muscular dystrophy, Pompe disease, and certain myosinopathies. In addition, metabolic and chronic disorders such as obesity, type 2 diabetes, heart failure, chronic obstructive pulmonary disease, or aging-related sarcopenia affect certain fiber types, while other fiber types seem to be resistant [15] (Table 5.1). The capacity of the muscle to alter the characteristics of its fibers may provide beneficial effects in the prevention and treatment of these diseases [10, 15].

Energy Substrates

ATP

Skeletal muscles store a relatively small amount of ATP which can support muscle contractions for only a few seconds. No differences in ATP concentration in different fiber types of human skeletal muscle have been observed [4, 16, 17]. A decline in muscle ATP concentrations is associated with muscle fatigue. Muscle fatigue is a protective mechanism designed to prevent ATP decline to levels associated with muscle rigor or serious muscle damage [18, 19].

Phosphocreatine (PCr)

Skeletal muscle PCr reserves are about three times higher than ATP levels. Its function is to replenish ATP via rephosphorylation of ADP. PCr stores are higher in type II compared to type I fibers [17, 20, 21]. Furthermore, PCr content may be increased by dietary manipulation and in

Table 5.1 Muscle disorders and morphological and functional changes of affected fiber types

Morphological/functional change	Fiber type affected	Muscle disorder
Atrophy-degeneration	Type IIx	Duchenne muscular dystrophy
	Type I	Myotonic dystrophy type 1 Myosinopathies Muscle inactivity (injury, bed rest)
	Type II Type IIa	Myosinopathies Pompe disease (mouse model) Aging/sarcopenia
Fiber type shift	Type II → type I	Facioscapulohumeral muscular dystrophy Congenital fiber type disproportion Heart failure (diaphragm) Chronic obstructive pulmonary disease (diaphragm)
	Type I → type IIx	Obesity Type 2 diabetes Muscle inactivity (injury, bed rest)
	Type I→ type II	Heart failure (limb muscles) Chronic obstructive pulmonary disease (limb muscles)
Reduced force generation	Type I	Myotonic dystrophy type 1
	Type II	Facioscapulohumeral muscular dystrophy

Modified from Ref. [15]

particular creatine supplementation and exercise [22]. The outcome varies among individuals and seems to be affected by factors such as dietary habits (vegetarians vs. omnivorous) or age (children vs. elderly) [23].

Glycogen

Glycogen is the main form of carbohydrates used for muscular work. It is also the most advantageous energy fuel in terms of ATP resynthesis since glycogen degradation is accomplished both aerobically and anaerobically. Glycogen is a polymerized form of glucose stored mainly in muscle and liver tissue. Its structure is in branch form in a treelike formation. This arrangement provides an advantage to enzymes phosphorylase and transferase to rapidly reach the various terminal sides of glycogen formation and speed up glycogen breakdown, making glycolysis a very fast metabolic pathway. Similarly, the many end points of the treelike formation provide multiple sites to the glycogen synthase for glucose unit addition through the process of glycogenesis. Ultimately, glycogen, this important substrate, can be degraded and resynthesized quickly [24].

About 75% of glycogen is stored between myofibrils as inter-myofibrillar glycogen, while the rest of the total glycogen pool is situated in the myofibrils and beneath the sarcolemma (intra-myofibrillar and sub-sarcolemmal glycogen, respectively) [25, 26]. In healthy individuals, muscle glycogen concentration varies depending on the tissue, the preceding physical activity, the person's recent diet, fitness status, fiber type, and possibly gender [26–28]. The liver tissue can accommodate approximately 85 kg wet weight^{-1}, or approximately 100 g of glycogen for the average liver weighing 1.2 kg [24]. In recent years, the use of ^{13}C magnetic resonance spectroscopy studies has shown that liver glycogen content does not differ between trained and untrained individuals and declines significantly during submaximal endurance exercise of about 60–70% VO_2max [27, 29, 30]. Conversely, muscle glycogen levels are usually 20–66% higher in endurance trained compared with untrained individuals. This may be attributable to increased insulin sensitivity observed with exercise training [27]. Furthermore, muscle glycogen is higher in type II fiber types compared to type I [4, 6, 16, 31]. Muscle glycogen stored can be increased by manipulation in diet and exercise, the so-called carbohydrate loading strategy or supercompensa-

tion, often used by endurance athletes to improve performance [28]. Whether this supercompensation response differs between males and females still remains controversial [2, 32, 33].

Finally, the existence of several types of glycogen storage diseases caused by various enzyme deficiencies [34], although rare, produces metabolic abnormalities in the liver, muscle, and brain and is associated with abnormal glucose and fat metabolism.

Lipids

Lipids are stored mainly in the adipose tissue and muscle in the form of triacylglycerols. There is considerable variation among individuals in terms of total body fat stores that can exceed 50% of the total body weight in severely obese. In general, females have a higher body fat content than males, and sportsmen usually have lower body fat levels than inactive individuals, although there is also a great variability in fat weight among athletes [35].

Intramuscular triglycerides are stored in the form of lipid droplets close to the mitochondria. Their amount varies between muscle groups and between fiber types, with type I to have a higher content as reported in muscle biopsy and magnetic resonance spectroscopy studies [36, 37]. The quantification of intramuscular triglycerides and their contribution to energy metabolism is problematic due to the fact that these fat reserves are not as nicely distributed in the muscle as glycogen [38]. However, studies combining immunofluorescence microscopy, stable isotope, and muscle biopsy techniques have demonstrated that muscle triglycerides are important energy contributors during prolonged exercise (≥3 h) of low to moderate (approximately ≤60% VO_2max) intensity [39].

Metabolic Pathways

As mentioned previously the energy source for muscular work is ATP. For work to continue, a constant supply of ATP is needed. This is accomplished via the anaerobic and aerobic pathways.

Anaerobic Metabolism During Exercise

The ATP-PCr System

During the hydrolysis of ATP by myofibrillar ATPase, ADP, hydrogen ions, and inorganic phosphate are formed as well as 30.5 kj of free energy per mole of ATP [40]:

$$\mathrm{ATP} + \mathrm{H_2O} \leftrightarrow \mathrm{ADP} + \mathrm{Pi} + \mathrm{H^+} - 30.5\,\mathrm{kj}\left(\text{kilojoules}\right) \tag{5.1}$$

This energy release provides the "driving or power stroke" by which the myosin attachment at a 90° angle to the binding sites on actin to change to a 45° angle resulting in the shortening in the muscle [41]. For the detachment of myosin from actin to take place, ATP is also required to bind to myosin. So, ATP is regenerated by the conversion of ADP and inorganic phosphate, so that ATP is again available to myosin. The majority of the energy spent (about 70–75%) in the contracting muscle is used by the myosin ATPase activity, with the remaining amount to be used by enzymes involved in Na^+, K^+, and Ca^{2+} ATPase [42].

The amount of energy stored in the form of ATP is limited (about 25 $\mathrm{mmol.kg^{-1}dw}$) and, if not resynthesized, can only provide energy for approximately 3–5 s of sprinting or about 15 s of aerobic exercise [42]. One way of the anaerobic ATP provision is accomplished via the breakdown of PCr, a reaction catalyzed by creatine phosphokinase (CPK) or creatine kinase (CK), leading to the formation of ATP and creatine (Cr):

$$\mathrm{PCr} + \mathrm{ADP} + \mathrm{H^+} \leftrightarrow \mathrm{ATP} + \mathrm{Cr} \tag{5.2}$$

During maximal efforts lasting more than 10 s, ATP stores decline in the exercising muscle, while ADP and adenosine monophosphate (AMP) are formed. This allows the formation of some ATP by a reaction (5.3) catalyzed by adenylate kinase [42]:

$$2\mathrm{ADP} \leftrightarrow \mathrm{ATP} + \mathrm{AMP} \tag{5.3}$$

The produced AMP is quickly broken down by the enzyme AMP deaminase to inosine monophosphate (IMP) and ammonium (NH_4^+):

$$AMP + H^+ \rightarrow IMP + NH_4^+ \quad (5.4)$$

Since NH_4^+ is toxic, it is transported in the liver through blood circulation and converted to urea. The formation of IMP is important in maintaining ADP and AMP at low levels in the muscle cell sustaining in this way enough free energy from ATP breakdown to support muscle contraction [19].

The above three reactions (5.2, 5.3, and 5.4) formulate the "ATP-PCr system" [43], "the phosphagen system" [19], or "anaerobic alactic system" since no oxygen or lactate formation is involved [44]. As highlighted earlier, the importance of this system is that energy can be provided at a very high rate almost instantaneously, something that cannot be accomplished by glycolysis or aerobic metabolism. This system has played a crucial role in our survival, as immediate action is required in many instances to avoid injury. Activities that require high levels of muscle power (weightlifting, shot put, hammer and discus throw, jumping, etc.) are also supported by the phosphagen system.

Glycolysis

ATP generated via glycolysis involves ten chemical reactions with lactate as the end product formed by pyruvate, a reaction catalyzed by lactate dehydrogenase (LDH). The net result for the muscle is the formation of 2ATP molecules when G-6-P is derived from glucose and three molecules when the initial substrate is glycogen. This pathway can be summarized as follows:

$$\text{Glycogen} + 3ADP + 3Pi \rightarrow 3ATP + 2\text{Lactate} + 2H^+ \quad (5.5)$$

A central reaction in glycolytic pathway is considered the transformation of 3-phosphogly ceraldehyde to 1,3-diphosphoglycerate by glyceraldehyde phosphate dehydrogenase, where nicotinamide adenine dinucleotide (NAD^+) is reduced to NADH [40]. The ratio of NAD/NADH, the so-called redox (reduction-oxidation) status of the muscle, plays a significant role as substrate in electron transport chain and oxidative phosphorylation since for every pair of electrons transported by NADH to the electron transport chain, three molecules of ATP are generated. Furthermore, NAD and NADH have the role of activators or inhibitors in muscle metabolism [42].

Although not as fast as ATP-PCr system, glycolysis can also regenerate ATP very quickly and is activated within the very first second of muscle contraction by various factors such as Ca^{2+}, ADP, AMP, IMP, fructose-6-phosphate, Pi, and Mg^{2+} in an allosteric fashion [44]. It can support sporting activities that require high level of ATP resynthesis for several seconds such as sprinting during various games like soccer, basketball, football, handball, and several others. Furthermore, glycolysis provides the extra energy needed when intensity exceeds that of VO_2max (Fig.5.4) or when oxygen is not available. In addition, both the phosphagen system and glycolysis support ATP regeneration at the initiation of exercise (Fig. 5.1) as well as when exercise intensity changes to a higher intensity level, allowing the necessary time for the aerobic pathways to match the ATP demands (Fig. 5.3).

Regulation of Anaerobic Pathways

The regulation of anaerobic pathways is mainly achieved by activation or inhibition of the enzymes catalyzing the various reactions. With the initiation of muscle contractions, the accompanied increase in Ca^{2+}stimulates myofibrillar ATPase. This stimulation moves ATP hydrolysis reaction (5.1) to the right, resulting in a decrease in ATP concentrations and a subsequent elevation of ADP and Pi. These changes produce a mass-action effect stimulating CPK to displace reaction (5.2) to the right replacing the previously hydrolyzed ATP but "spending" PCr stores [45]. It should be noted that as mentioned earlier, although ATP concentrations do not massively change even in maximal fatiguing exercise, small changes of this molecule have a larger impact on the concentrations of ADP and AMP which together with ATP formulate the total adenylate pool [46].

The enzyme phosphofructokinase (PFK) responsible for the conversion of fructose-6-phosphate to fructose-1,6-diphosphate is

regarded as the key enzyme in the control of glycolysis [46]. In activities like sprinting, the increase in the rate of glycolysis exceeds 1000-fold compared to resting rate [40]. Increased cellular levels of inorganic phosphate (5.1), as a result of ATP hydrolysis, as well as AMP by adenylate kinase (5.3), enhance PFK activity. On the other hand, when ATP demand is not high and ATP levels rise, PFK activity is reduced. A high rate of glycolysis will also result in an increased H^+ accumulation (5.5) that will lower muscle pH inhibiting in this way PFK. This is considered a protective mechanism, designed to prevent further lactate formation leading to extreme acidosis [46]. ATP activity is also inhibited by increased citrate concentrations, an intermediate of Krebs cycle, indicating that when ATP is regenerated by aerobic metabolism there is no need for high rates of ATP formation through glycolysis.

In muscle glycogenolysis, the main control is exerted by glycogen phosphorylase (PHOS) that degrades glycogen to G1P (Fig. 5.1). This enzyme has two forms: PHOS *a*, the more active, and PHOS *b*, the less active which are interconvertible by phosphatase and kinase enzymes. When the muscle is at rest, most of PHOS is in its less active *b* form [40]. Activators of PHOS from *b* to *a* form are ADP, AMP, IMP, Pi, Ca^{2+}, and adrenaline, whereas inhibitors are H^+, G6P, and ATP [44, 47, 48]. Also, the availability of substrates in the form of exogenous oral carbohydrates, blood-borne fatty acids, or muscle glycogen itself may influence the rate of muscle glycogenolysis [6, 31, 49–52].

Lactate Metabolism

Lactate is the end product of glycolysis formed after pyruvate is reduced to lactate by LDH according to the following chemical reaction:

$$\text{Pyruvate} + \text{NADH} + \text{H}^+ \leftrightarrow \text{Lactate} + \text{NAD}^+$$

Often the term lactic acid is used instead of lactate and vice versa. Lactic acid, when formed, is unstable within the physiological muscle and blood pH range and immediately more than 99% of lactic acid releases a proton and dissociates into lactate anions and protons (H+) [53]. Therefore, since lactate is measured, this term will be used in this chapter. In biochemical terms, the above reaction is important because lactate regenerates NAD in the cytosol that is necessary in the glyceraldehyde phosphate dehydrogenase which in turn converts 3-phosphoglyceraldehyde to 1,3-diphosphoglycerate. In this way the glycolytic flux and redox status of the muscle (NAD/NADH) are maintained; otherwise, glycolysis would slow down resulting in a reduced glycolytic rate of ATP resynthesis. This becomes very important not so much during high-intensity exercise, where PCr stores are significantly reduced within seconds and consequently the muscle relies more on glycolysis, but in endurance events such as marathon running. Fast marathoners rely mainly on carbohydrate oxidation during exercise; however, this requires a high rate of glycogenolysis and simultaneously the ability to remove and metabolize lactate for energy [54, 55].

Two misconceptions exist regarding lactate. First, lactate is associated with metabolic acidosis and fatigue. However, the lactate molecule itself is not responsible for acidosis. In reality, the production of lactate coincides with the formation of H^+ that reduces pH-inhibiting key enzymes of glycolysis such as PFK [56]. In fact, to some extent, lactate contributes to proton buffering since in the LDH reaction (conversion of pyruvate to lactate), two electrons and one proton from NADH and another proton from solution are used [19].

Second, especially in the past, lactate was considered a waste product of glycolysis due to hypoxia and was associated with fatigue especially during high-intensity exercise [57]. Also, postexercise lactate metabolism was linked to the oxygen depth, the phenomenon of elevated oxygen uptake during the postexercise period [58]. However, since the 1970s, the multidimensional metabolic role of lactate was realized gradually [53, 57]. The application of muscle biopsy; arteriovenous, magnetic resonance spectroscopy; and tracer techniques enabled researchers to focus not simply on lactate accumulation during or after exercise but on lactate formation (appearance), removal, and transport between and within various tissues such as the muscle, liver, heart, and brain [59–61]. For example aerobically

trained individuals demonstrate a delayed blood lactate accumulation during progressive exercise compared to anaerobic or untrained people, as indicated by the higher anaerobic threshold they possess [62], suggesting that lactate formation and removal can be modulated by exercise training. This is discussed later on in this chapter.

Currently, the general consensus is that lactate formation is not due to hypoxic conditions of the working muscles since an increased lactate production and accumulation also occurs under aerobic conditions [53, 57].

The concept that lactate produced by the working muscles is taken up and metabolized by other tissues was introduced in the mid-1980s by George A. Brooks and was termed lactate shuttle, although today is known as cell-to-cell lactate shuttle [63]. He reported that about 75% of the lactate during submaximal exercise is removed and oxidized and only about 20% is converted to glucose. Lactate continues to be oxidized during the recovery period, but significant amounts are used for glycogen repletion through gluconeogenesis in the muscle and liver. However, these tissues do not replace their glycogen via this mechanism unless feeding is provided, with the exception of the heart that supercompensates its glycogen stores even during fasting [63]. The cell-to-cell shuttle has been proposed not only for the muscle but other tissues like the brain, heart, and liver.

Lactate can also be transported to oxidative (type I) fibers within the exercising muscle, to inactive oxidative fibers, and used as energy source. It can also exit the working muscle and be transported to less active muscle groups or to inactive glycolytic (type II) muscle fibers where it is either oxidized or converted to glycogen and stored [53, 57]. The liver can also oxidize or convert lactate to glucose, where this newly formed glucose can be either returned to exercising muscle through the Cori cycle pathway or stored as liver glycogen. Also, lactate in the systemic circulation can be directly oxidized in heart and brain tissue. Regarding the brain, astrocytes take-up blood glucose and convert it to lactate that in turn is transported to nearby neurons where it can be oxidized to regenerate ATP in the mitochondria [53, 57]. Therefore, taking into account all these possible pathways, lactate today is considered as a metabolic intermediate that connects glycolytic with oxidative metabolism [54].

In medicine, lactate metabolism has recently being incorporated in the study of cancer metabolism and treatment [64]. On the basis of Warburg effect, where in the presence of oxygen, cancer cells rely on glycolysis for energy production accompanied by high rates of lactate formation, it has been suggested that a treatment approach would have been to neglect glucose to cancer cells and provide as alternative fuel lactate, the so-called lactate-protected hypoglycemia treatment [65]. Although this idea is attractive, cancer metabolism is far more complicated with a diverse collection of normal and cancer cells whose metabolism is different in an in vitro environment compared to poorly understood tumor microenvironment [53, 64]. Nevertheless, it seems that as our understanding on lactate metabolism increases, this molecule will have the potential to offer more in health and disease in the future.

Lactate Threshold: How Is It Affected by Fitness?

In 1964 Wasserman and McIlroy introduced the term "anaerobic threshold" (AT) when attempting to define (without assessing blood lactate) the exercise intensity where energy production shifted from a mainly aerobic metabolism to that combining both anaerobic and aerobic patterns [66].

When VO_2max is directly assessed, ventilatory rates (V_E; volume of air inspired or expired per minute), volume of carbon dioxide expired (VCO_2), VCO_2/VO_2 ratio referred as the respiratory exchange ratio (RER or simply R), and the ventilatory equivalent for oxygen (VO_2/V_E) are also assessed. As the workload increases, a point is reached where these values increase disproportionally to exercise intensity [67]. This reflects the point at which the increasing metabolic demands due to the increased workload can no longer be met mainly by aerobic metabolism. Consequently, an increase in the relative contribution to energy needs is met by anaerobic

metabolism and especially by glycolysis. This point has been coined as the "anaerobic or ventilator threshold."

This exercise level is also marked by an increase in lactate concentration accompanied by an increase in H^+ which is eventually transported out of the muscle in the circulation and blood pH is reduced. Thus, this threshold can also be detected using blood lactate measurements and is therefore referred to as lactate threshold (LT). An example of such determination is presented in Fig. 5.5. Because the AT and LT do not always coincide and, at times, the term onset of blood lactate accumulation (OBLA) has been introduced, in this case, the LT is defined as the exercise intensity at which a lactate accumulation of 4 $mmol.l^{-1}$ is observed [68]. As illustrated in Fig. 5.5, this blood lactate level occurs at 9 $km{\cdot}h^{-1}$. Below this workload, lactate remains fairly constant despite an increase in exercise intensity and an expected increase in glycolysis (Fig. 5.5). This indicates that lactate formed below workload of 9 $km.h^{-1}$ is metabolized by the working muscle (via the cell-to-cell shuttles discussed above), and therefore lactate concentrations do not rise until the 9 $km.h^{-1}$ workload is exceeded.

Since the original study by Wasserman and McIlroy in 1964, much criticism and debate has taken place, mainly regarding the link between what is observed and the actual changes occurring within the exercising muscle. This is due to methodological constraints and the difficulty in measuring intracellular partial pressure of oxygen during incremental exercise [56]. However, LT and AT still remain valuable functional parameters for doctors and sports scientists [69–71].

Can the Lactate Threshold Be Altered?

The LA is determined by mainly two factors: genetics and the level of physical fitness. Since there is little one can do with our genetic make-up, AT can only be altered by physical activity. The change in AT occurs in two ways. First, regularly performed aerobic activities lead to an increased VO_2max. The increase in VO_2max dictates that O_2 consumption at a given submaximal workload will be lower postexercise training compared to pre-training. As an example, let us assume that VO_2max prior to exercise training was 40 $ml^{-1}.kg^{-1}.min^{-1}$ and following training increased to 50 $ml^{-1}.kg\text{-}^{1}.min^{-1}$. If we assume that LT occurs at approximately 50% of VO_2max, the AT for this individual prior to exercise training will occur at 20 $ml^{-1}.kg^{-1}.min^{-1}$ and at 25 $ml^{-1}.kg^{-1}.min^{-1}$ after training. Thus, following exercise training, the workload necessary to shift the working muscle from predominantly aerobic to anaerobic metabolism is increased.

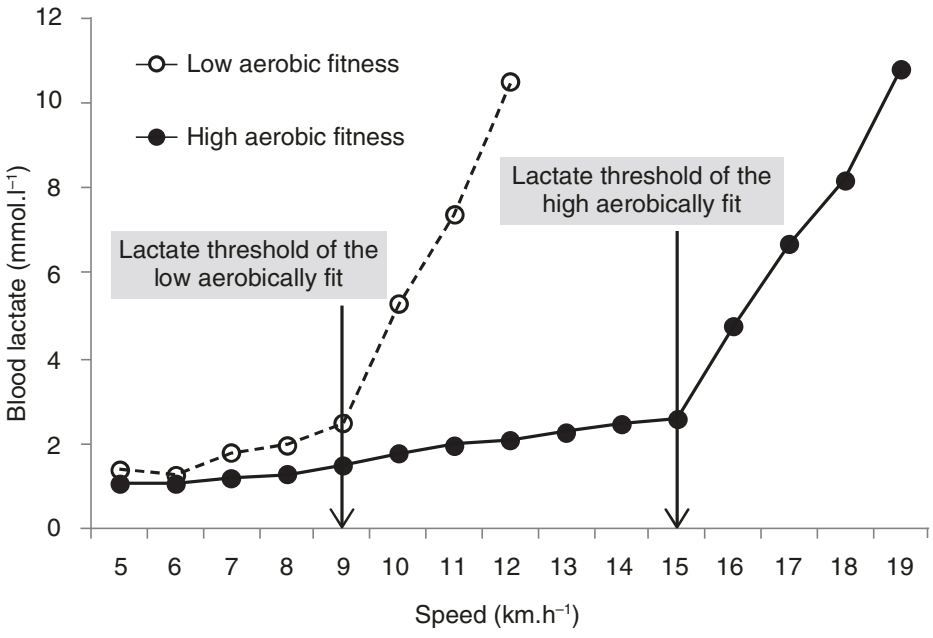


Fig. 5.5 A hypothetical lactate threshold during grated exercise test to volitional fatigue for low and high aerobically trained individuals

The second way AT is altered is via the increased efficiency of the aerobic pathways resulting from training. In other words, the metabolic alterations in aerobic enzymes such as succinate dehydrogenase, malate dehydrogenase, citrate synthase, a higher number and size of mitochondria, improved capillary network, and a high percentage of type I muscle fibers [68, 69, 72, 73] render the aerobically trained muscle capable of exercising at higher exercise intensities and accumulating even less blood lactate [74]. Thus, the AT of highly trained individuals occurs at about 75% and not at 50% as is the case with sedentary individuals. This is illustrated in Fig. 5.5 where an individual with low aerobic fitness may have a LT at 9 $km^{-1} \cdot h^{-1}$, while for someone with high aerobic fitness, LT can occur at 15 $Km^{-1} \cdot h^{-1}$.

Aerobic Metabolism During Exercise

Aerobic metabolism involves more complex energy systems than anaerobic metabolism, since all three major macronutrients, carbohydrates, fats, and proteins, contribute to the production of ATP. All aerobic chemical reactions take place in the mitochondria, situated mostly near the myofibrils, but may also be scattered in the sarcoplasm. A common molecule of all macronutrients is acetyl-Coa, although some other metabolites from fat or protein origin may "join" the oxidative phosphorylation process at glycolysis or Krebs cycle pathways.

The main characteristic of this system is that it can provide a much higher amount of ATP to the working muscle and for fats this may be almost unlimited, but its major disadvantage is that this energy is given at a substantially lower rate compared to anaerobic metabolism. For example, the net ATP yield of PCr system is 1 ATP, and glucose/glycogen 2–3 ATP. In contrast, the complete aerobic oxidation of glucose/glycogen yields 38–39 ATP, and a fatty acid like palmitate per mol 129 ATP. The maximum rates of ATP resynthesis by the aforementioned systems are approximately 2.25, 1.10, and 0.25–0.70 $ATP \cdot kg^{-1}$ $ww.s^{-1}$, for PCr, glycolysis, and Krebs cycle, respectively [40, 75].

Carbohydrate Metabolism

Blood-borne glucose and muscle glycogen are the carbohydrate substrates converted to pyruvate and then enter mitochondria as acetyl-CoA (Fig. 5.1). This reaction is catalyzed by the pyruvate dehydrogenase complex (PDH), situated in the inner mitochondrial membrane. PDH consists of three enzymes: pyruvate dehydrogenase, dihydrolipoamide acetyltransferase, and dihydrolipoamide reductase [40]. This chemical reaction is important since acetyl-CoA is the main substrate for the Krebs cycle. Once acetyl-CoA is formed, it cannot be converted back to pyruvate. Therefore, PDH is controlled by hormones and various effectors allosterically [40].

The Krebs cycle, also known as tricarboxylic acid cycle (TCA) or citric acid cycle, consists of a series of reactions that begins with the combination of oxaloacetate and acetyl-CoA to form citrate. These reactions are summarized as follows [40]:

$$\text{Acetyl-CoA} + 3H_2 0 \rightarrow 2CO_2 + 4[2H] + \text{CoASH}(\text{Coenzyme A})$$

Each cycle generates four pairs of hydrogen atoms ([2H]) which are carried to the electron transport chain by NADH and flavin adenine dinucleotide ($FADH_2$). Through the aerobic process of oxidative phosphorylation, the complete oxidation of acetyl-CoA yields 12 ATP molecules per cycle. A complete oxidation of glucose yields a total of 38 ATP, and glycogen yields 39 ATP. This is 19 and 13 times higher, respectively, compared to 2 and 3 ATP generated anaerobically.

Regulation of Hepatic Glucose Production

Liver plays a dominant role in blood homeostasis during exercise by releasing glucose into the bloodstream; otherwise, exercise would have been impossible to be carried out [76]. Splanchnic glucose increases almost linearly to exercise intensities up to 60% VO_2max and exponentially above this level, despite a gradual decrease in blood flow to the hepatosplanchnic area due to blood redistribution favoring the

exercising muscles [77]. This exponential increase of blood glucose at higher exercise intensities shows that hepatic glucose production (HGP) and glucose uptake by the muscle do not match, indicating that regulation of HGP during exercise may not be via a feedback mechanism (i.e., blood glucose concentration) [77]. This mismatch suggests that hormonal and neural factors may affect to at least some extent HGP during exercise. In a study using somatostatin to modulate glucagon and insulin, during exercise at low intensity (40% VO_2max) for 2 h, the absence of glucagon totally abolished HGP, while in the absence of insulin, there was a threefold increase in HGP compared to rest [78]. There is evidence to suggest that these two hormones and especially glucagon may be more important in maintaining blood homeostasis late into a long-duration exercise rather than at the beginning [79].

The fact that epinephrine stimulates glycogen degradation during exercise led to the assumption that epinephrine stimulates HGP during exercise [76]. However, collective findings from human and animal studies show that when sympathoadrenergic activity is reduced, splanchnic glucose production is not impaired [77]. This leads to the conclusion that in addition to glucagon, insulin, or epinephrine, other mechanisms may be involved in the regulation of HGP during exercise [76, 77].

Regulation of Muscle Glucose Uptake

Glucose diffuses from capillaries to the muscle membrane through interstitial fluid by facilitated diffusion and is converted to G-6-P by hexokinase. Thus, blood supply, transport, and phosphorylation inside the muscle cell are potential sites of regulation of glucose uptake by the exercising muscle [80]. Simply, glucose uptake increases with increasing exercise intensity and duration to support the continuous energy demands of exercising muscle [81–83]. In the classical studies by Ahlborg and co-workers, when blood glucose gradually declined to about 2.5–3 mmol.l^{-1} over 3.5–4 h of cycling, muscle glucose uptake was also reduced [81, 82]. On the other hand, when exogenous carbohydrate was provided during cycling, hypoglycemia was prevented and glucose uptake was maintained [84, 85]. These finding support that blood glucose concentration is an important factor for muscle glucose uptake during exercise.

The diffusion of glucose into the muscle cell is facilitated by the insulin-mediated translocation of GLUT-4 transporters from intracellular storage depots to the sarcolemma and transverse tubules. It is also well accepted that exercise has an insulin-like effect, translocating GLUT-4 to the surface of the cell through different molecular mechanisms and independent from insulin [80, 86]. Various factors identified as potential activators of GLUT-4 include Ca^{2+}, AMP-activated protein kinase (AMPK), reactive oxygen species (ROS), nitric oxide (NO), soluble *N*-ethylmaleimide-sensitive factor attachment protein receptors (SNARE), and GTPases especially RabGTPase proteins (members of the Ras small GTPases superfamily) [86–89]. Furthermore, epinephrine seems to reduce glucose uptake possibly due to stimulation of muscle glycogenolysis, resulting in an elevated G-6-P concentration. In turn hexokinase, the enzyme responsible for glucose phosphorylation, is inhibited [90]. Finally, lowering muscle glycogen content by 60 min single-legged cycling 16 h before a subsequent two-legged exercise bout increased muscle glucose uptake by threefold in the exercising leg (lower glycogen leg) compared to the control leg, suggesting that muscle glycogen levels may influence muscle glucose uptake [91].

Cross Talk Between Skeletal Muscle and Liver Metabolism in Exercise

During muscle contractions, skeletal muscle releases cytokines referred to as myokines [92]. In fact, the muscle is considered an endocrine organ that releases various myokines such as interleukin (IL) IL-6, IL-8, IL-15, brain-derived neurotrophic factor, leukemia inhibitory factor, fibroblast growth factor 21, and follistatin-like 1 [93]. In particular, muscle-derived IL-6, among other actions, may act in a hormonelike fashion increasing HGP and lipolysis in adipose tissue [94, 95]. In a study involving cycling at 40% VO_2max for 2 h with high and low IL-6 infusion

rates, the use of stable 6,6$^2H_2$ isotopes demonstrated higher rates of appearance and disappearance of blood glucose when the IL-6 infusion rates were high. This supports the view that IL-6 influences glucose homeostasis during exercise [95]. Furthermore, muscle-derived IL-6 has been observed to stimulate liver CXL-1 chemokine expression, a small cytokine that is involved in the processes of angiogenesis, inflammation, and wound healing, in exercising mice [96]. Thus, it is theorized that the exercising muscle communicates with distant organs such as the liver, adipose tissue, and brain by the release of myokines. This muscle-to-organ cross talk may play a significant role in the prevention of metabolic-related diseases such as obesity, diabetes, and cancer [93, 97].

Fat Metabolism

Acetyl-CoA is also generated from fat metabolism. Fats in the form of triacylglycerols are degraded to fatty acids and glycerol by a hormone-sensitive lipase through the process of lipolysis. Once in the circulation, glycerol is either taken by the liver to form triacylglycerols, converted to glucose through gluconeogenesis, or converted to dihydroxyacetone phosphate and enters glycolysis.

Fatty acids in the circulation are bound to albumin, whereas fatty acids in the muscle are also bound to binding proteins (Fig. 5.1). When fatty acids enter the muscle cells, they are converted to fatty acyl-CoA, which, in turn with the aid of carnitine, crosses the mitochondrial matrix. This is catalyzed by carnitine acyltransferase, also named carnitine palmitoyltransferase. This enzyme exists in two forms, one bound to the outer mitochondrial membrane (carnitine acyltransferase I) producing acyl carnitine and the other on the inner mitochondrial membrane that reverses the previous reaction producing acyl-CoA and carnitine [46]. Inside the mitochondrion, fatty acyl-CoA gradually loses two carbons to form acetyl-CoA, which enters Krebs cycle following the same pathway pyruvate follows (Fig. 5.1). Fatty acids yield more ATP depending on the number of their carbon atoms. For example, a 16-atom fatty acid will generate a net yield of 130 ATP molecules. Due to the complexity of mobilization, transport, and b-oxidation processes, however, the rate of ATP resynthesis by fatty acids is the slowest among all the fuels available [75].

Regulation of Muscle Fat Metabolism

The potential sites for regulation in skeletal muscle fat metabolism are (a) lipolysis of the adipose tissue and the delivery of fatty acids to the muscle, (b) movement of fatty acids across sarcolemma, (c) control of these molecules through the mitochondrial membrane, and (d) regulation of triacylglycerol lipase activity [98].

The hormone-sensitive lipase (HSL) responsible for lipolysis is activated by catecholamines which through β-adrenergic receptors activate lipolytic cascade and HSL is phosphorylated [99]. When participants cycled for 30 min at three different exercise intensities, 25%, 65%, and 85% VO_2max, fatty acid uptake, and oxidation decreased with the highest exercise intensity [100]. The investigators concluded that this was the outcome of reduced blood flow to the adipose tissue. However, in a subsequent study where fatty acid concentration was maintained high at exercise levels of 85% VO_2max, the uptake and oxidation of fatty acids increased, but was still lower than the levels achieved when exercising at 65% VO_2max. This suggests that intramuscular factors may be responsible for the rate of fat oxidation at higher exercise intensities [101].

Regarding the movement of fatty acids across muscle membrane during exercise, three fatty acid transport proteins, fatty acid binding protein in plasma membrane ($FABP_{pm}$), the fatty acid translocase (FAT/CD36), and fatty acid transport protein (FATP) have attracted considerable attention [102, 103]. In particular, evidence supports that FAT/CD36 is translocated from intracellular space to the muscle membrane during muscle contraction, and this coincides with increased fatty acid transport to the muscle [98]. Furthermore, more recent observations suggest that FAT/CD36 translocation is activated by insulin. This transporter is also present in the subcellular and intra-myofibrillar mitochondria, and its mitochondrial content is correlated with mitochondrial fatty acid oxidation,

indicating that it is involved in regulating fatty acid oxidation in human skeletal muscle [103–105]. Another potential regulatory factor in fat oxidation is the muscle triacylglycerol lipase (MTGL). Methodological constraints have not allowed MTGL to be studied thoroughly. However, it seems that it is activated during aerobic exercise and contributes to energy supply by the degradation of muscle triacylglycerols which play an important role in providing fatty acids for oxidation during exercise [39, 98, 103].

Proteins as an Energy Source During Exercise

As mentioned earlier the protein contribution to ATP resynthesis is minimum. The muscle has the capacity to oxidize seven amino acids (alanine, asparagines, aspartate, glutamate, isoleucine, leucine, and valine), with some evidence suggesting that lysine may also be oxidized [106]. However, during exercise the main amino acids oxidized are isoleucine, leucine, and valine, the so-called branched-chain amino acids (BCAA). This is indicated by isotope tracer studies, a large muscle uptake of BCAA, and specific enzyme activation [107]. The BCAA are first transaminated to their keto acid analogues by branched-chain aminotransferase, and the formulated keto acids are oxidized by a mitochondrial branched-chain 2-oxoacid dehydrogenase enzyme (BCOADH) [106]. The BCOADH is considered to be the rate-limiting enzyme for BCAA oxidation and exists in active dephosphorylated form and less active phosphorylated form [106, 107]. This enzyme is activated during endurance exercise in human muscle [108–110]. This activation seems to be related to the glycogen status in the muscle, since when glycogen stores are low BCOADH is activated more, whereas when glycogen stores are high, the enzyme is not activated [110, 111]. This is linked to the observation that based on sweat urea N measurements low carbohydrate stores elevate protein degradation during exercise [112]. However, the contribution of BCAA to total energy expenditure during endurance activities is calculated to be only about 3–6% [1, 2]. Nevertheless, there is the view that amino acids may interact with TCA during prolonged exercise. As exercise progresses and glycogen stores become depleted, an increase in leucine oxidation may lead to a carbon drain on the TCA cycle, reduce its flux, and lead to fatigue [3, 111]. However, this mechanism may not be quantitatively important by others since changes in TCA cycle intermediates may be unrelated to oxidative energy provision in skeletal muscle [113].

Finally, in resistance exercise there is an increased protein turnover in the postexercise period that remains negative (i.e., protein breakdown >protein resynthesis) if the exercising individual remains in postabsorptive condition [107, 114].

Factors Determining Substrate Preference During Exercise

Substrate preference for ATP resynthesis in healthy individuals is dependent on four main factors, exercise intensity, duration, fitness status, and diet, while gender and environmental factors may also influence substrate utilization. The contribution of PCr is mainly at exercise intensities exceeding 100% VO_2max or when exercise intensity increases abruptly (Figs. 5.3 and 5.4). Therefore, the main fuels used are carbohydrates and fats, while proteins contribute approximately <5–6% to the total energy turnover [1, 2].

Possibly the strongest factor dictating the relative contribution of fats and carbohydrates is exercise intensity. At low exercise intensity levels of about 25% VO_2max plasma fatty acids are the main contributors. As intensity increases, the contribution of fats decreases proportionally, with a marked decrease observed at exercise intensities above 80% of VO_2max, while the contribution of muscle glycogen and plasma glucose increases proportionally [100, 115]. However, fat oxidation in absolute amounts (i.e., g/min) increases up to exercise intensities of 40–65% VO_2max [115, 116]. It seems that an increased glycolytic flux may inhibit fatty acids from entering the mitochondria, leading to diminished lipid oxidation [117, 118]. However, since all the aforementioned studies were based on indirect calorimetry, there

is the concern that at high exercise intensities, fat oxidation might have been underestimated due to metabolic perturbations [119].

Another major factor that alters substrate preference is the duration of exercise. In general, at fixed exercise intensity, fat oxidation increases, while carbohydrate contribution decreases as exercise progresses. At low to moderate exercise intensities (about 25–60% VO_2max), fats gradually become the main fuels for energy especially when activity lasts for hours. For example, in one of the classical studies by Ahlborg and coworkers, they reported that during 4 h of exercise at approximately 30% of VO_2max the relative contribution of free fatty acid rose progressively to 62% of the total energy requirements after 40 min of exercise. The contribution of glucose fell from 40% during the initial exercise of 40 min to 30% between 90 and 240 min [81].

Similarly, in another study, plasma fatty acids provided more than 80% of total energy expenditure during 2 h of cycling at 25% VO_2max [100]. Finally, Edwards and colleagues reported that a runner exercising for 6 hours, about 84% of the energy requirements are derived from fats [120]. The increased free fatty acid (FFA) oxidation over time is associated with a progressive decrease in glycogen stores lending support to the concept that carbohydrate availability may play a role in the regulation of fat oxidation during exercise [117].

It is also well documented that aerobic fitness status and diet influence substrate use. This occurs as a result of metabolic adaptations occurring at peripheral fat tissues and within the muscle cells enabling the trained muscle to oxidize relatively more fat and less carbohydrate [121–123]. This is of particular importance for the endurance athlete since the limiting factor in performance is glycogen stores. Shifting the balance to utilizing more fat than glycogen for a given task (marathon run) delays glycogen depletion and, therefore, delays fatigue [56].

The effect of diet on substrate use was demonstrated almost a century ago when Krogh and Lindhard showed that after a high-fat low-carbohydrate diet RER values were lower from baseline, indicating higher oxidation of FFA [124]. More recent studies confirmed this early finding and reported that a high-carbohydrate diet reduces fat oxidation, whereas a diet rich in fat produces the opposite result [125]. Similarly, carbohydrate oxidation increased during exercise after a carbohydrate load 3–4 h prior to exercise compared to an overnight fast [126–128]. It takes at least 6 h at the postabsorptive state before exercise metabolic responses are similar to a fast of 8–12 h [129].

Finally, environmental conditions may play a role in substrate use especially when these conditions divert from a thermoneutral environment. In general, when exercise is performed in the heat, there is an increase in muscle glycogenolysis and liver glucose output, reduced fat utilization, and also an enhanced and no increase in muscle glucose uptake, leading to an elevation in blood glucose concentration. These responses seem to be related to high muscle temperature and a sympathoadrenal response [130].

Sex-Related Differences in Substrate Preference During Exercise

It is well established that sex-related metabolic and hormonal differences in exercise exist and should be considered [131, 132]. Males rely to a greater extent on carbohydrate oxidation, while females rely more on lipid oxidation displaying a lower RER during exercise. Indeed, at any given relative exercise intensity, women oxidize less carbohydrate and more fat than men [133, 134]. However, other studies reported no gender differences in the substrate preference [115, 135]. The rationale for the possible increased fat oxidation by females may be that estrogen promotes fat oxidation, as indicated when estrogen supplementation is administered in males [136]. Female hormones or the relatively greater proportion of type I fibers in women compared to men may explain the differences between the patterns of energy substrate utilization in men and women [132, 137]. In addition, elevated levels of GH in women, both at rest and at peak response to exercise, could also contribute to the sex-related differences in substrate use [132]. Furthermore, variations in ovarian hormone levels throughout

the menstrual cycle may alter exercise metabolism in women [138].

Specifically, the female sex hormone, estrogen, is a factor that in both sexes affects energy substrate selection during exercise; however, it does not appear to be the sole determinant for substrate selection in females [139]. Estrogen, specifically 17β-estradiol, decreases carbohydrate oxidation and promotes lipid oxidation during prolonged, moderate-intensity exercise [140–142]. Moreover, during the luteal phase compared to the follicular phase of the menstrual cycle, higher levels of circulating estrogen and thus a higher relative rate of fat oxidation as well as a higher estradiol response to an acute bout of exercise have been observed [132, 143, 144]. In addition, a greater reliance on lipids as a fuel source was found in females compared to males during prolonged, moderate-intensity exercise, indicated by a lower RER in women. Moreover, women in the follicular phase had higher glycogen use as well as glucose appearance and disappearance rates than women in the luteal phase [141]. Interestingly, during the follicular phase of the menstrual cycle, women exhibit an increase in circulating estradiol after an acute bout of aerobic or resistance exercise, in contrast with men, although circulating levels of estrogen do not differ significantly between women and men [143, 145, 146]. Moreover, there is evidence supporting that overall fat oxidation may not be different between sexes, but the type of fat may differ with females to use more myocellular triacylglycerols [147, 148]. The issue becomes more complex due to the anti-estrogenic action of progesterone and other confounding variables such as exercise intensity and nutritional status that may obscure any estrogen's possible effect [138].

Cardiac Metabolism During Exercise

Myocardial metabolism is a complex network of highly regulated metabolic pathways and an integral part of the function of the heart, both as consumer and provider of energy, matching cardiac energy demand and supply with precision [149]. Cardiac metabolism in health and disease has been studied extensively in vitro, in animal models and in humans under resting conditions [150–153]. However, few studies in vivo have been conducted in healthy humans during exercise. This is due to technical and practical difficulties as well as to the invasive nature of these procedures, where the methods commonly used are catheterization of aorta and coronary sinus and use of radioactive and nonradioactive isotopes [154].

Metabolism of Energy Substrate in Heart During Exercise

The basic principles of cellular metabolism in various tissues apply also to cardiac muscle cells, with some quantitative differences. The cardiac muscle is designed to function aerobically, as it is endowed by an abundance of blood supply. Under resting conditions, most of the energy requirements of the heart are derived from fatty acids.

During exercise, cardiac output may increase more than sevenfold exceeding 40 $l \cdot min^{-1}$ in elite endurance athletes [11]. Therefore, the heart muscle requires large amounts of energy and is the largest energy consumer relative to its weight in the body [155]. Since the ATP content of the heart muscle is low (5 $\mu mol \cdot g^{-1}$ww), ATP homeostasis must be maintained for myocardium to function properly [151]. To support ATP resynthesis, the myocardium uses a variety of substrates such as fatty acids, glucose, lactate, pyruvate, ketones, and amino acids [155]. However, the main substrates that have been directly studied and seem to contribute during exercise to the oxygen extraction ratio of the myocardium are free fatty acids, glucose, and lactate [156–162]. During exercise, the preferred fuel for the cardiac muscle is lactate released form the exercising muscle followed by FFA. However, under ischemic conditions (i.e., coronary artery obstruction), the heart is forced to switch to anaerobic glycolysis for its energy needs. This situation is not sustainable, and if blood supply is not restored within minutes, the heart suffers irreparable damage. The relation between energy supply, mechanical function, and intracellular pH of the myocardium, both in

healthy and diseased conditions, has been fundamental to cardiology.

Glucose

Glucose utilization for the myocardium occurs especially under hypoxic conditions, resulting in lactate formation [159, 162]. Multiple functions in myocardium are also mediated by glucose and its metabolites, and failure to control the levels of intracellular glucose metabolites has been implicated in the generation of ROS, as well as the development of insulin resistance. Moreover, excessive accumulation of glucose metabolites has been associated with various myocardial pathologies [149].

Free Fatty Acids

Long-chain free fatty acids are the myocardium's predominant fuel for respiration [163]. The pathway of long-chain FFA metabolism (oxidation) is initiated with their liberation from triglycerides (TGs) and ends with the entry of acetyl-CoA into the Krebs cycle. Acetyl-CoA is committed to oxidation by the system of β-oxidation, inside the mitochondria [164]. The rate of oxidation of FFA in the myocardium is somehow related to the activity of Krebs cycle and the rate of oxidative phosphorylation, and changes in flux through pathways of fatty acid metabolism reflect changes in substrate provision to the myocardium [149].

Ketone Bodies

Concentrations of ketone bodies in the plasma can dramatically rise during exercise, and their uptake, which is concentration-dependent, is a key feature in myocardial metabolism. The ketone bodies have access to the Krebs cycle in the heart [165]; however, the rate of oxidation is not sufficient to meet the energy demands of the myocardium [166, 167]. Interestingly, pyruvate carboxylation accounts for at least 3–6% of Krebs cycle flux in the heart [149].

Amino Acids

An integral part of energy metabolism in the heart is also amino acids, and one of the main functions of transaminases in myocardium under physiologic conditions is the supply of carbon skeletons for the Krebs cycle [168]. In addition, alanine is also an end product of anaerobic glucose metabolism, like lactate, and it is easily transphosphorylated to ATP, while this reaction results in anaerobic energy production independently of lactate formation [149].

As in skeletal muscle and other tissues, most of the metabolic energy is used to form ATP in the mitochondria, and this ATP acts as the conveyer of energy for cardiac muscle contraction and other cellular functions, such as ion movements and intracellular protein turnover. As in any other bodily organ, it is impossible to separate metabolism from function in the heart. The heart converts substrates and oxygen to contractile function and heat, and there is positive correlation between the work output, the rate of ATP turnover, the rate of oxygen consumption, and the rate of substrate input and utilization. However, it is certain that for a given physiologic environment, the myocardium oxidizes the most efficient substrate [149].

Preferred Substrate for Energy Needs of the Heart During Exercise

Metabolism of oxidizable substrates for energy supply fuels ATP production in the mitochondria for the contractile function of the myocardium. About 2/3 of the energy from the hydrolyzed ATP is used in contractile machinery with the remaining to support ion pumps such as Ca^{2+}, Na^{+}, and K^{+}. The main driving force of energy metabolism in cardiac muscle is the rate of energy turnover and not its stored ATP [149, 169–171].

The intermediary metabolism of energy substrates supports the contractile function of the myocardium, and the bulk of the energy for this function is derived from oxidative phosphorylation of ADP. Indeed, in well-oxygenated healthy myocardium, ATP resynthesis is accomplished almost exclusively (>98%) by oxidative phosphorylation in the mitochondria, and only a small fraction (<2%) is derived from glycolysis [172]. Interestingly, the myocardium has the capability of maintaining and controlling, even during high-intensity exercise, the same levels of

high-energy phosphate compounds (PCr and ATP) as well as their ratio observed at rest, suggesting that the intracellular-free ADP concentration, differently from skeletal muscle, does not function as a primary system to control cardiac muscle during exercise [173, 174]. Indeed, using ^{31}P NMR technology in vivo it was observed that even a threefold increase in myocardial oxygen did not alter cytosolic ATP, ADP, and Pi concentrations [175].

In particular, the total lactate dehydrogenase (LDH) and phosphofructokinase (PFK) activity is high, and, therefore, cardiac muscle should have the ability to release significant energy via anaerobic glycolysis [176, 177]. However, the myocardium also has a high aerobic capacity, and, therefore, both at rest and during maximal exercise, myocardial energy demands are met mostly by aerobic metabolism without detectable contribution of anaerobic glycolysis, even during maximal exercise [178, 179]. Notably, in a healthy heart the coronary circulation has the capacity to supply the myocardium with blood and oxygen without the need of anaerobic myocardial metabolism [176]. Indeed, aerobic metabolism is predominant in myocardium and the predominant fuel for energy supply is FFA. Glucose is not a preferred substrate by the cardiac muscle, and when it is limitedly used, myocardium first oxidizes glycogen, followed by the aerobic, again, breakdown of glucose and lactate, without the need of anaerobic metabolism. Interestingly, during high-intensity exercise, when blood levels of lactate rise, the healthy preferred substrate for the myocardium is lactate, which replaces all other energy-providing substrates as heart's fuel for respiration [149, 180–183].

Molecular Mechanisms of Exercise-Induced Metabolic Adaptations in Cardiac Muscle

Exercise-induced physiologic cardiac adaptations include mitochondrial biogenesis, ultimately leading to enhanced fatty acid and glucose metabolism. Consequently, metabolic homeostasis is preserved [179]. Metabolite profiling before and after exercise showed a subset of metabolites that regulate glucose and lipid metabolism[184], suggesting that metabolites and other molecules regulate various physiological processes, possibly including the myocardium response to exercise [185].

Moreover, sirtuins (SIRTs), a family of NAD-dependent deacetylases, regulate a variety of functions in the cells, including growth, metabolism, apoptosis, and aging [186]. SIRT1 and SIRT3 are the most studied in the cardiac tissue and both are upregulated by exercise. SIRT1 has pro-growth and pro-survival functions in cardiac muscle cells [187], while SIRT3 is a mitochondrial sirtuin [188] and protects the heart against oxidative stress. It has also been reported that it may modulate the opening of the mitochondrial permeability transition pore (mPTP) [189, 190]. Additionally, it regulates cardiac metabolism via activation of 5' AMP-activated protein kinase (AMPK) and peroxisome proliferator-activated receptor gamma coactivator 1-alpha (PGC-1α), both of which inhibit maladaptive remodeling of myocardium [189].

Furthermore, exercise upregulates PGC-1α, as potent mediator of oxidative phosphorylation and mitochondrial biogenesis. PGC-1α-deficiency results in inability of the myocardium to meet energy demands, indicating the importance of energy and metabolic homeostasis in myocardial health [179]. PGC-1α has also been shown to regulate a pathway of angiogenesis which is independent of the hypoxia-inducible factor-1 (HIF-1), therefore providing a mechanism for coordinately regulating blood supply and mitochondrial function in the exercising myocardium [185, 191].

Future Considerations of Cardiac Metabolism

Both in physiological and pathological conditions, the cardiac muscle has enormous energy demands, and myocardial metabolic dysregulation is a noticeable feature of cardiovascular disease [185]. The importance of energy substrate

metabolism in the myocardium is increasingly appreciated in heart disease, diabetes, and cancer. Changes in the physiologic environment of the heart directly result in alterations of its metabolic fluxes, suggesting that an integral part of cardiac adaptation to its environment is metabolic signals, while metabolic remodeling both triggers and sustains structural and functional remodeling of the heart [192]. Notably, pathological cardiac remodeling has been associated with a switch from the fatty acid utilization, which is the primary energy substrate for the adult heart, to glucose metabolism, as it happens in ischemic conditions. Since aerobic exercise training has been shown to promote not only efficient fatty acid and glucose handling but also mitochondrial biogenesis in the heart [179], aerobic exercise benefits in myocardial metabolic dysregulation should be further characterized and utilized, as they cannot be fully reproduced by any novel and improved, exercise-mimicking treatment for heart disease [185].

Exercise-Induced Chronic Metabolic Adaptations

Regularly performed exercises of adequate intensity, duration, and volume lead to chronic adaptations. These adaptations are specific to the exercise mode, encompass both morphological and function changes, and include all systems involved in the physical task. The ultimate outcome is improved functional capacity of the individual resulting in an improved oxidative capacity of the working muscle [26, 193–197]. A major metabolic change due to endurance training is the exercise-induced mitochondrial biogenesis resulting in a higher mitochondrial content, volume, and oxidative capacity [198].

This process involves multiple molecular events ultimately resulting in improved functional capacity of the individual [198, 199] and significant health implications, as these adaptations attenuate the aged-induced sarcopenia and apoptosis that can lead to pathological conditions such as autoimmune and neurodegenerative diseases [199].

The increase in the number, volume, and oxidative capacity of mitochondria is accompanied by increased mitochondrial enzyme concentration and activity and increases in capillary density [197, 200, 201]. Collectively, these adaptations increase the capacity of the muscle to oxidize fats and carbohydrates more efficiently, sparing in this way the limited body glycogen stores [56]. This is significant for athletes engaged in long-duration activities (marathon run), where one of the limiting factors of performance is glycogen depletion leading to fatigue.

Significant reduction in the rate of muscle glycogenolysis with endurance training has been reported in several studies [200, 202–204]. However, others dispute these findings, as change in the rate of muscle glycogenolysis is not accompanied by a higher activation of phosphorylase, indicating that other factors such as ADP, AMP, and Pi may have an influence [203]. Furthermore, studies using isotopes have reported a reduced rate of liver glycogenolysis as indicated by the lower rate of glucose appearance in the systemic circulation [204–206]. Conflicting results have also been reported regarding gluconeogenesis [205, 207]. Muscle glucose uptake is also reduced as a result of training at pre-training exercise intensity of 60% VO_2max [206, 208, 209]. However, at higher pre-training exercise intensities (80–100% VO_2max), post training muscle glucose uptake is increased [210].

Studies have shown clearly that fat oxidation is enhanced with aerobic training [206–209]. However, it is difficult to clearly distinguish fat oxidation from blood-borne fatty acids with that from intramuscular stores. Methodological constraints in arterial-venous difference and tracer procedures have resulted in conflicting results regarding the extent to which blood-borne fatty acid oxidation is elevated [211]. It is not clear if there are exercise-related improvements in intramuscular triglycerides oxidation. Accurate assessments are hampered by the variability of biopsy samples as well as the potential that some of the fatty acids from circulation may not be oxidized in the muscle but replace intramuscular fat stores [211]. However, it is the discrepancy between total fat oxidized by indirect calorimetry

and that from tracer data which suggests that intramuscular fatty acids are oxidized to a greater extent after training [211, 212].

Another clear training response is the lower lactate concentrations after aerobic training. This reflects an enhanced metabolic clearance rate as judged by the rate of lactate disappearance at any given concentration [59, 213, 214]. The precise mechanism(s) facilitating lactate clearance is not understood. However, several concepts are proposed including (1) enhanced mitochondrial oxidative capacity, (2) an increase in the expression of monocarboxylate transporter 1 (MCT1) which facilitates lactate uptake by mitochondria, and (3) alteration of LDH activity to its H-LDH isoenzyme that favors lactate oxidation to pyruvate [215].

Hormonal Regulation of Exercise Metabolism

Hormones are sensitive to exercise-induced stress and play various roles in muscle metabolism during exercise or even in the regenerative and adaptive mechanisms following exercise-induced muscle damage [44, 137, 216].

Specifically, hormones regulate in part the release of energy from carbohydrate and lipid stores during exercise, the synthesis of glycogen and TGs following meals, and the resynthesis of muscle protein. Within the context of energy storage and energy production during exercise, the role of catecholamines, insulin, glucagon, and cortisol in the metabolic regulation of carbohydrate and lipid metabolism will be considered, along with the mechanisms that elicit the metabolic responses. Moreover, as exercise is accompanied by changes in catabolic and anabolic hormones, as well as muscle protein synthesis, the roles of growth hormone (GH), testosterone, and estrogen will also be described.

The increased energy demands during exercise are modulated by the synergistical work of pancreatic hormones, insulin and glucagon, resulting in elevated blood glucose [137, 217, 218]. Specifically, during exercise, insulin and the contractile activity of the exercising muscles act synergistically, but through independent mechanisms, to facilitate translocation of glucose transporter type 4 (GLUT4) receptors to the cell membrane and increase glucose uptake of the exercising muscles [137, 219, 220]. Although glucose transporters on plasma membranes are activated by both insulin and exercise independently [221–226], insulin inhibits the transcriptional biosynthesis of GLUT4, and, thus, it leads to a marked reduction of the GLUT4 pool of the low-density microsomes (LDM), where GLUT4 is stored. However, exercise not only increases the GLUT4 translocation from LDM compartment to the plasma membrane but also increases the biosynthesis of GLUT4, thus leading to only a small decrease of GLUT4 pool in LDM [221, 227–229]. Interestingly, in fat cells only insulin, and not exercise, accelerates the GLUT4 translocation velocity from LDM to the plasma membrane [221, 230].

Long-term muscular activity during prolonged moderate exercise results in greater insulin sensitivity, and, thus, blood insulin levels decrease [231–233]. Thus, during an ultra-long distance running, a rise of insulin antagonists, i.e., cortisol, glucagon, and GH, has been observed in the early stages of ultra-long running, which was inversely proportional to the intake of carbohydrates [234]. During endurance exercise, there are a number of potential sites of control which can regulate the interaction of substrate (carbohydrate and lipid) metabolism. These include availability of intra- and extra-muscular substrates which are controlled by diet and the action of key hormones, such as insulin and the catecholamines, the abundance of proteins such as GLUT4, and the activity of key enzymes involved in the regulation of metabolic pathways. Thus, interestingly, within the context of interactions between hormonal actions and metabolic regulation, insulin, apart from acting to lower blood glucose, it may also stimulate glycolysis by inducing increased synthesis of key glycolytic enzymes [44].

In addition to insulin and glucagon, blood glucose during exercise is also modulated by cortisol and epinephrine. Exercise-induced stress results in the release of both these hormones, and both mediate the maintenance of blood glucose levels.

Specifically, epinephrine, a stress hormone secreted by the adrenal medulla, stimulates glycogen phosphorylase activity in the liver and muscle, thereby increasing glucose availability for the exercising muscles. Cortisol, also a stress hormone, is secreted from the adrenal gland and promotes muscle protein breakdown, thus making certain glucogenic amino acids available for gluconeogenesis (after 20–30 min or more) and ultimately resulting in increased blood glucose levels [137].

In summary, exercise facilitates an increase in circulating cortisol, catecholamines, glucagon, and GH and a suppression of insulin release. Consequently, these hormones synergistically increase glycolysis, glycogenolysis both in the muscle and liver, lipolysis in muscle and adipose tissue, gluconeogenesis in the liver, and protein degradation in the muscle and liver. Ultimately, this leads to an increase in substrate availability to meet the increased energy demands of the working muscles.

In addition to the effects of the hormones on energy availability during exercise, it is also important to appreciate that hormones regulate the recovery process following exercise. This includes not only resynthesis of muscle glycogen stores ready for the next training session but also promotion of protein synthesis in the muscle. For instance, increased release of GH in response to exercise may have an anabolic effect or improve postexercise recovery time [44].

Hormonal Responses in High-Intensity Anaerobic Exercise and in Prolonged Aerobic Exercise

Repeated high-intensity (near maximal or supramaximal) exercise bouts of activity lasting only seconds to a few minutes, interspersed with exercise of low to moderate intensity (active recovery) or complete inactivity, have become popular recently. The major sources of energy during such activities are derived from anaerobic processes. On the other hand, aerobic processes provide the energy sources of physical activity that is performed for durations from a few minutes to hours and causes an increased heart rate and respiratory volume to meet the oxygen requirements of the exercising muscles [44, 235, 236].

A growing interest has been developed regarding the metabolic characteristics and adaptations of high-intensity exercise training [237]. In this context, the acute hormonal and metabolic responses to high-intensity anaerobic exercise have received particular interest. More specifically, high metabolic stimuli, such as high levels of lactate, changes in the acid base status, and large decreases in pH, which are induced by high-intensity interval training (HIIT), elicit certain hormonal responses that may be different from the traditional anaerobic work and appear to result even in aerobic adaptations [238–240]. Metabolic disturbances appear to play a key role in causing acute hormonal responses and long-term adaptations to HIIT, and it has been speculated that the acute metabolic disturbances and the consequent hormonal increases after HIIT may play a positive role in optimizing training adaptations and eliciting health benefits. The hormonal responses to HIIT indicate their involvement in adaptation mechanisms potentially as part of a regulatory network to support a normal adaptation process to HIIT. However, the influence of the duration of intervals and recovery, the different exercise intensities, and the work/rest ratio on the acute hormonal responses to HIIT and particularly on its long-term adaptations should be further investigated and optimized [237]. Moreover, it remains to be elucidated if these responses and adaptations have significant health implications, such as the improvement of some cardiometabolic risk factors reported in special populations [241].

Exercise represents a powerful stimulus for the sympathoadrenergic system, which is important for the metabolic adaptation to exercise [217, 242, 243]. Specifically, during exercise the stimulation and the subsequent increase in plasma catecholamines depend on the duration of exercise (total work), workload, and the muscle mass recruited, as well as on the emotional stress, however, not on the power output pattern, in both continuous and interval exercise training. Particularly during high-intensity exercise, the

sympathoadrenergic system and catecholamines affect the substrate mobilization, while particularly noradrenaline is a potent stimulator of muscle glycolysis [137]. Moreover, although the concentration of blood glucose strongly influences the extent of the insulin secretion, however, insulin release is inhibited by stimulation of adrenoceptors on pancreatic beta-cells, as it occurs during exhaustive exercise due to the increased levels of catecholamines. Increases in catecholamine concentrations are higher in intense anaerobic exercise than prolonged aerobic exercise in both young and adults [137, 244, 245]. The adrenergic inhibition of the insulin secretion by the exercise-induced elevated catecholamine levels supports the substrate supply during exercise; glycogenolysis and lipolysis are inhibited by insulin, while, in contrast, catecholamines that oppose the action of insulin stimulate these processes [137]. Thus, the inhibition of insulin release results in reduced glycogenesis in liver and muscle and intensifies glycogen mobilization in muscle and the glycogenolytic and gluconeogenetic glucose output from the liver [217]. In addition, within the context of hormonal-induced substrate mobilization, repeated bouts of high-intensity sprints have been shown to result in increased blood levels of glucagon, with no effect of sex on those changes [246]. Nevertheless, exercise training-induced changes in hormone concentrations such as norepinephrine, insulin, and glucagon are unable to explain all of the effects that occur between liver glucose production and muscle glucose uptake during exercise, and it has been proposed that, possibly, the actual rate of muscle glucose uptake acts as a feedback signal to regulate glucose output from the liver [44, 209].

Growth hormone possesses both anabolic and catabolic actions. It stimulates cellular uptake of amino acids and their incorporation into various proteins, including those of the muscle. It also acts as a repartitioning agent fostering fat metabolism via mobilization of TGs. In adults, GH levels increase during exercise; however, its secretion is pulsatile in nature, and, thus, it is difficult to interpret its peak values during exercise [106, 137, 247, 248]. There is evidence that GH responses are much higher in HIIT compared to high-volume aerobic training and that the increase in blood GH levels is in part a consequence of the decreased blood pH following HIIT [240].

It is important to mention that, when examining GH response to exercise, insulin-like growth factor-1 (IGF-1) should be taken into consideration. IGF-1 is a hormone that mediates many actions of GH, and the GH-IGF-1 axis, among other primary actions, has been suggested to mediate many of the anabolic effects associated with resistance, anaerobic, and aerobic exercise [249, 250]. Specifically, during exercise IGF-1 levels appear to be independent of GH responses, while there has been an inconsistency of findings regarding the IGF-1 response to exercise, with studies reporting a decrease, increase, or no change in blood IGF-1 [251, 252]. Most studies reported significant increases in IGF-1 after high-intensity exercise stimuli, while IGF-1 response to exercise appears to depend on type, intensity, and duration of exercise. Particularly regarding IGF-1 and glucose response, it is not clear to what extent endogenously produced IGF-1 contributes to glucose homeostasis, although there is evidence that exogenous IGF-1 can lower blood glucose [132].

Serum levels of hormones such as GH, IGF-1, testosterone, estradiol, dehydroepiandrosterone, and cortisol have been found to be similarly increased in response to an acute bout of moderate endurance exercise in adult females of a wide range of age, indicating that increasing age does not necessarily inhibit the hormonal response to a bout of aerobic exercise in women [145]. In addition, the effect of exercise training on the hormonal responses of GH, IGF-1, testosterone, free testosterone (FT), and sex hormone-binding globulin (SHBG) to a sub-maximum aerobic exercise bout was investigated in older men before and after 4 months of resistance or moderate aerobic training. Aerobic training or leg-only resistance training did not change the resting hormonal concentration of older men. There was an increase in testosterone and FT concentration immediately after both sub-maximum aerobic exercise and resistance exercise bout, which was

higher after the 4-month resistance training but not after the aerobic training. In contrast, GH/IGF-1 response to sub-maximum aerobic exercise bout appeared to be blunted regardless of training status [253].

Gender Differences in Hormonal Responses in Anaerobic and Aerobic Exercise

Significantly higher GH responses in men compared to women after aerobic, anaerobic, or resistance exercise have been reported [132]. Moreover, females and males show a different pattern of GH release in the circulation during exercise, which peaks sooner and return to baseline more quickly in women, while men exhibit a more prolonged response. There are also noticeable sex-related differences in GH levels at rest, and subsequently higher peaks of GH during exercise in women. These differences in GH response have been attributed to a lack of testosterone response to exercise in women. Testosterone is a steroid hormone with anabolic potential on a number of tissues, including muscle and, hence, can impact muscle growth and exercise performance. Indeed, data suggest that this hormone may affect both anaerobic and aerobic performance [254, 255], with greater increases in FT being reported after HIIT than endurance exercise [256]. Women exhibit little or no increase in circulating testosterone in response to exercise [132, 257], and, hence, GH may compensate for the anabolic requirements triggered by acute exercise. In addition, the higher resting basal level of GH in women compared to men is dependent on the phase of the menstrual cycle, when estrogen levels affect accordingly circulating GH levels (reviewed in [132]). The sex-associated differences in GH response to exercise can affect the control of blood glucose in both sexes; increases in GH stimulate lipolysis and lipid oxidation while suppressing glucose oxidation and, consequently, increasing blood glucose levels [257]. Thus, higher levels of GH at rest in women, due to higher levels of estrogen, may preserve blood glucose levels to a greater extent in women compared with men [132, 257].

Nevertheless, the majority of studies investigating the GH/IGF-1 axis responses to exercise reported a similar relative increase in both sexes during and after exercise longer than 10 min [132, 257–259], while slight decreases in IGF-1 responses to ultra-endurance exercise were revealed, again similarly occurring in both sexes [132, 260]. Acute bouts of HIIT have been shown to lead or not to significant differences in IGF-1 responses in male compared to females, while the increase in IGF-1 in response to HIIT does not appear to be depended on the phase of the menstrual cycle of the female subjects [132, 250].

Moreover, it was showed that long-duration, low-intensity, or moderate aerobic exercise bouts produced significantly lower epinephrine and norepinephrine levels in women than in men, while there is a sex-related difference in sensitivity of lipolytic activity to catecholamines during exercise. Catecholamines increase lipolysis during exercise, and despite their lower levels in women compared to men, there has been evidence for elevated levels of lipolysis in women during exercise, implying a greater sensitivity to the lipolytic action of the catecholamines in females. Thus, during exercise-induced elevation of epinephrine, women have relatively greater fat oxidation and lipolysis than men [132, 259]. In addition, blood glucagon increases in both sexes following prolonged submaximal exercise, with the majority of studies reporting a lower glucagon response to moderate exercise in females compared to males [132, 259, 261].

Exercise Metabolism: Clinical Implications

Diabetes Mellitus (Type 1 and 2)

Diabetes mellitus refers to a group of metabolic disorders which are characterized by chronically increased circulating glucose levels (hyperglycemia). There are two main forms of diabetes, type 1 (T1D) and type 2 (T2D). T1D usually begins in youth, also known as juvenile-onset diabetes, whereas T2D usually begins in adulthood, and thus it is referred to as adult-onset diabetes [262].

Hyperglycemia is a common feature of both forms of diabetes; however, the cause is different. In T1D an autoimmune destruction of the beta-cells in the pancreas that produce insulin results in no insulin production in most patients (a minority of T1D patients has some remaining β-cell function). These patients require exogenous insulin, and for this reason, this type of diabetes mellitus is also known as insulin-dependent. Its etiology remains unknown; however, environmental factors, genetic disposition, and autoimmune reactions have been implicated [263]. Exogenous insulin use has been greatly contributed to the prevention of hyperglycemia and management of T1D. Exercise has also been shown to be effective in the management of hyperglycemia by improving glucose uptake form of the exercising muscles [137, 219, 221–226, 264].

T2D, accounting for up to 95% of all cases, is characterized by insulin resistance, in which the response to insulin in the muscle, liver, and fat cells is inadequate. However, endogenous production of insulin in many instances is also impaired. T2D, also known as non-insulin-dependent diabetes mellitus, as exogenous administration of insulin may not be necessary, and insulin resistance can be managed using diet and exercise to enhance insulin sensitivity [262]. Since insulin is required for muscle cells, liver cells, and adipocytes to take up and store glucose [262, 265], insulin resistance and insulin deficiency lead to hyperglycemia. Hyperglycemia is associated with blood vessel wall and nerve damage, eventually leading to various complications and premature mortality, usually from cardiovascular disease [266–271]. Moreover, increased physical activity and enhanced cardiorespiratory fitness are well accepted as an effective approach to attenuate and even prevent the development of T2D in individuals with prediabetes [266, 269, 272].

Diabetes Mellitus and Exercise

The improved glucose uptake by exercise, due to increased GLUT4 density in plasma membrane, has therapeutic consequences for both T1D and T2D. Specifically, both acute exercise and especially chronic exercise training lead to an insulin-independent increase in glucose uptake by GLUT4. This partially compensates for the absence of the insulin-stimulated glucose uptake in T1D. In T2D, characterized by insulin receptor and post-receptor defects and insulin resistance, acute and chronic exercise not only improves GLUT4 function but also enhances B-cell function and improvement of insulin sensitivity [217, 273, 274].

It is not clear whether there are sex-related differences in insulin sensitivity in response to exercise, since some studies have reported that women show a greater improvement of insulin sensitivity in response to acute bouts of submaximal exercise, while, conversely, others have shown that men and women exhibit a similar improvement of insulin sensitivity [132]. Furthermore, the phase of the menstrual cycle may also affect insulin sensitivity during exercise in women, as a significant decrease in insulin sensitivity was found during the luteal phase compared to the follicular phase (reviewed in [132]).

Type 1 Diabetes Mellitus and Exercise

The findings on the specific effects of exercise training on the glycemic control of patients with T1D are conflicting. Some reported no improvement in glycosylated hemoglobin (HbA1c) concentrations with physical training, whereas an inverse association between physical activity levels and HbA1c in T1D patients has been reported in a comprehensive review [263]. Moreover, the Position Statement of the American Diabetes Association for physical activity/exercise and diabetes [275] states that regular physical activity/exercise has an important role in the treatment of T1D and related benefits, including improved long-term weight and glycemic control, insulin sensitivity, lipid profile, and endothelial function, as well as fitness level and overall well-being [263, 264, 275–279].

Individuals with T1D who take up exercise have specific needs, as both exercise and insulin therapy play a key role in glycemic control. Since exercise enhances glucose uptake, exogenous insulin administration (injection) and carbohydrate ingestion must be coordinated with exercise. Specifically, when to exercise in relations to

the meal and/or insulin injection, how hard (intensity), and how long (duration) are important consideration to maintain glycemic control [276]. Failure to appropriately coordinate blood glucose, dietary needs, and exogenous insulin administration prior, during, or following exercise can lead to unfavorable health outcomes [280, 281].

In particular, the increased release of counter-regulatory hormones to provide with energy for exercise of longer duration increases the risk of hypoglycemia particularly after endurance exercise. This risk persists for up to 15 h after the completion of exercise. It has been reported that overnight hypoglycemia is common in children with T1D after exercise, underlining the importance of blood glucose management in these patients when exercise is performed afternoon [262].

Various exercise strategies, including a single, all-out 10-s sprint before or after a bout of prolonged, moderate aerobic exercise, have been developed for the prevention of postexercise hypoglycemia in individuals with T1D. Indeed, there is evidence that high-intensity exercise has a stronger impact on glycemic control than moderate exercise. The risk of hypoglycemia is also lower with HIIT compared with moderate-intensity continuous training, as HIIT stimulates glucose production in the liver more than moderate-intensity exercise. Thus, athletes with T1D must be instructed so as to avoid exercise-induced hypoglycemia, by monitoring blood glucose and adjusting insulin and diet [263, 282].

Overall, although balancing exercise and food intake and adjusting insulin dosage are a challenge individualized for the T1D athlete in order to prevent either hyperglycemia or hypoglycemia, it is attainable, and physical activity/exercise recommendations should be adapted to the specific needs of each individual with T1D. Exercise training must be regular and in accordance with insulin treatment and adjustment, as well as with dietary regulation [263, 275, 276, 278, 280, 281, 283]. Both aerobic exercise and strength training of moderate-intensity exercise are advisable, as well as their combination, for at least 30 min daily. Exercise training has been recommended to be of approximately the same intensity and, if possible, at the same time of day [263]. Moreover, while exercise is safe and beneficial for individuals with T1D and avoiding exercise carries greater risks than being active, however, in order to reduce the risk of some complications, precautions must be taken. Thus, it has been suggested that physical activity should be postponed when blood glucose levels are higher than 14 mmol/L accompanied by ketonuria, or higher than 17 mmol/L without ketonuria, until this condition has been corrected. The same applies to blood glucose levels lower than 7 mmol/L [263].

Type 2 Diabetes Mellitus and Exercise

T2D is a progressive disease in which insulin resistance leads to poor glycemic control (hyperglycemia), and since this condition is not due to the lack of insulin, the body continues to secrete more insulin in response to the hyperglycemic state, resulting in hyperinsulinemia [262, 276]. There is a large body of evidence that physical exercise reduces the risk of developing T2D, while, also, exercise is one of the three cornerstones in the treatment of T2D along with healthy diet and medication [262, 263, 276, 277]. Specifically, many studies have indicated that regular physical activity plays a key role in controlling blood glucose in patients with T2D. In particular, a meta-analysis revealed that exercise lowers postprandial glucose but not fasting glucose in T2D, and this is important because, unlike medications, exercise is effective in reducing postprandial glycemic excursions just within a few days [263, 284]. Moreover, exercise improves glucose tolerance, insulin sensitivity, and HbA1c, while muscle contraction triggers glucose transport by insulin-independent mechanisms [262]. Indeed, physical exercise leads to an increase in insulin sensitivity and, consequently, in glucose uptake in insulin-sensitive tissues, however, with a lower consumption of insulin. Thus, the aforementioned long-term effect of exercise on glycemic levels can be expected [263]. A detailed coverage of the topic is provided by the Position Statement of the American Diabetes Association for physical activity/exercise and diabetes, briefly discussed below [275], and the American College of Sports Medicine (ACSM) and the American

Diabetes Association Joint Position Statement for Exercise and T2D [285].

Observational and intervention studies, randomized controlled trials, and meta-analyses of controlled clinical trials concerning the effects of physical activity on T2D have shown health benefits of regular physical activity/exercise for the treatment of T2D. In particular, it has been revealed that aerobic exercise training significantly reduces HbA1c levels in individuals with T2D, while strength training also increases insulin-mediated glucose uptake in skeletal muscle and significantly decreases HbA1c in patients with T2D. Interestingly, no differences have been reported between aerobic training and resistance training regarding the effect on HbA1c changes [263, 286]. Specifically, progressive resistance training was found to be effective in improving insulin sensitivity in both children and adults. Indeed, strengthening skeletal muscle is strongly associated with improved glycemic control, since approximately 85% of glucose uptake takes place directly in skeletal muscle [276, 287]. In addition, HIIT improves glycemic control and induces cardiometabolic adaptations similar to those of moderate aerobic exercise in prediabetes and T2D, while it provides greater benefits to functional capacity in patients with T2D [263, 286, 288]. On the other hand, a clear advantage of various activities of moderate-intensity exercise lasting for 20–60 min per day to HIIT exercise for diabetes prevention was not identified [286]. Nevertheless, structured exercise training consisted of aerobic exercise, resistance training, or their combination was revealed by a meta-analysis to be associated with HbA1c reduction in patients with T2D. Interestingly, a combination of resistance training and aerobic training is probably the optimal form of exercise for patients with T2D. Furthermore, structured exercise training of more than 150 min/week has been associated with greater HbA1c improvements compared with that of 150 min or less/week [263, 289].

Overall, the Position Statement of the American Diabetes Association (2016) provides a clinically oriented review and evidence-based recommendations about physical activity and exercise in people with diabetes. Briefly, exercise improves blood glucose control in T2D, contributes to weight loss, improves well-being, and reduces cardiovascular risk factors. Moreover, regular exercise may prevent or delay the development of T2D and also has considerable health benefits for people with T1D. Blood glucose management through exercise includes various challenges related to diabetes type, activity type, and presence of diabetes-related complications [275]. In view of the optimum exercise prescription for reducing the risk of T2D, initially 150 min per week of moderate-intensity physical activity and building up to 200–300 min per week was proven effective in improving insulin sensitivity and, thus, can help in preventing T2D. More specifically, exercise recommendations include low- to moderate-intensity exercise (40–70% of maximum oxygen uptake) performed on at least three non-consecutive days each week, starting with 10–15 min and progressing up to 60 min per session over time [262]. The mode of exercise may depend on personal preference and include a variety of activities, comprised of aerobic and resistance exercises. More studies are needed to establish specific guidelines regarding the independent and synergistic effects of quantity and intensity of the various types of exercise [263, 285]. Although the health benefits of physical activity/exercise outweigh the risks, exercise should be postponed in T2D patients when blood glucose levels are >17 mmol/L or <7 mmol/L, until they have been corrected [263].

An important determinant of T2D risk is obesity, and although obesity and physical inactivity are both independent predictors of T2D risk, the power of this association is much greater for obesity compared with physical inactivity [262]. Indeed, obesity plays a pivotal role in the pathogenesis of insulin resistance in skeletal muscle; nevertheless, exercise represents one of the most effective interventions for reversing insulin resistance in skeletal muscle of obese patients at high risk for T2D [290]. Due to the strong association between physical activity, obesity, and T2D, there is currently great interest in these areas, as these conditions are related in part to a general decline in physical activity, while several new hormones discovered have enhanced understanding the mechanisms underlying diabetes and obesity [262]. Severe metabolic dysregulation in

diabetes and obesity can reduce the benefit from exercise; however, the intact response of key metabolic regulators in exercising muscle of diabetic patients indicates the effectiveness of exercise to treat these diseases [291].

Dyslipidemia-Hyperlipidemia

Hyperlipidemia or dyslipidemia is a group of disorders of lipid and lipoprotein metabolism characterized by increased circulating levels of certain forms of cholesterol and TGs. Isolated hypercholesterolemia and combined dyslipidemia are the most common types of dyslipidemia, occurring as a result of excessive fat intake and leading to increased risk of atherosclerosis [263, 292]. Regular exercise favorably modulates blood lipid profile and is considered as one of the mechanisms responsible at least in part, for the protective effects of exercise against the development of vascular diseases [280]. Moreover, epidemiological and cross-sectional observational studies indicate that physical activity prevents the development of hyperlipidemia [293–295].

Specifically, a systematic review assessing the effect of supervised exercise interventions on lipid profiles in patients with T2D concluded that exercise is effective in lowering low-density lipoprotein cholesterol (LDL-C) and elevating high-density lipoprotein cholesterol (HDL-C) levels in diabetic patients [263, 296]. There is also evidence supporting that a large volume of exercise training resulted in a beneficial effect on the blood lipid profile independently of weight loss [297]. Collectively, it is concluded that regular physical activity reduces TG and increases HDL-C levels in the blood [298, 299]. The amount of exercise required for favorable changes in the lipoprotein-lipid profile is approximately 7 miles or more per week. A dose-response relationship between miles run per week, HDL-C, and other lipoprotein-lipid levels was noted with most changes occurring when running 7–14 miles per week at mild to moderate intensities [300]. These findings are confirmed by an interventional study demonstrating that high-volume exercise had a more favorable impact on lipoprotein-lipid metabolism than exercise intensity [301].

Studies have also shown that exercise has favorable effects on postprandial lipid profile. Non-fasting TG levels were reduced significantly following exercise training in individuals with metabolic syndrome [302]. Moreover, a single exercise session appears to be as effective in lowering non-fasting TG as continuous aerobic exercise, with effects lasting till the following day [252, 303]. Similarly, short-term (4 days) aerobic exercise had effects on postprandial TG, LDL-C, and VLDL-C, but no changes in HDL-C were noted [304].

Collectively, the current literature suggests that aerobic exercise has a favorable effect on lipoprotein-lipid metabolism. An exercise intensity, duration, and volume threshold as well as an interaction between the exercise components (intensity, duration, frequency) appear to exist beyond which favorable changes can occur in a dose-response pattern [305, 306].

McArdle Disease

McArdle disease, also known as glycogen storage disease type V (GSDV) or myophosphorylase deficiency, is an inherited metabolic disorder characterized by the inability of skeletal muscle to degrade glycogen. Patients with McArdle disease are deficient in muscle glycogen phosphorylase [307–309]. Myophosphorylase is the only isoform of glycogen phosphorylase expressed in skeletal muscle only. Hence, McArdle disease is considered a relatively benign myopathy, as it affects only skeletal muscle in contrast with other metabolic disorders where, apart from muscle, other tissues and organs are also affected [310, 311].

The enzyme myophosphorylase is involved in muscle glycogen degradation to glucose-1-phosphate. Consequently, muscle phosphorylase deficiency renders the muscle incapable to mobilize and utilize muscle glycogen during an aerobic metabolism [312, 313]. Since glycolysis is blocked upstream, muscles can still take up and utilize blood glucose [311]; hence, glycolysis in skeletal muscles of McArdle disease patients is

not totally impaired. However, the substantially limited pyruvate formation generated from the limited glycolytic activity [314] leads to abnormally low substrate influx through the Krebs cycle and reduced rates of acetyl-CoA formation, thereby inhibiting the Krebs cycle oxidative phosphorylation [307, 311]. Consequently, VO_2max is approximately 40% lower than normal controls. This leads to a disproportionate elevation in exercise heart rate and ventilation rate which reduced blood flow to the exercising muscles, partial ischemia, and exacerbated symptoms [307, 311]. Muscle stiffness, fatigue, myalgia, and weakness, induced by exercise and relieved by rest, are also typical symptoms in these patients. If these symptoms are ignored and exercise is continued, painful cramping and contracture of the exercising muscles occurs, followed by myoglobinuria [307] and, in some cases, muscle damage or rhabdomyolysis [311, 314].

Free fatty acids are the primary energy substrate that is utilized by skeletal muscle, through oxidative phosphorylation, in the resting state as well as during low-intensity aerobic activity. Although acetyl-CoA is generated from free fatty acid metabolism, the capacity of the McArdle disease patients to utilize FFA without exercise training is limited [307, 313, 314]. Disorders that alter energy provision to the muscle, irrespective of whether they affect lipid or carbohydrate metabolism, essentially result in chronic muscle weakness or, most frequently in McArdle disease, exercise intolerance. Exercise intolerance is characterized by acute crises of muscle pain, stiffness, and undue fatigue, accompanied by muscle contractures, especially at the beginning of exercise, which are attenuated with exercise cessation; however, these crises can result in muscle damage or rhabdomyolysis [311, 314]. McArdle disease patients are likely to adapt a sedentary lifestyle exposing these patients to secondary health risks such as obesity, T2D, and cardiovascular disease [311, 315, 316].

Exercise Intervention Studies in McArdle's Disease

The exercise-related health benefits for McArdle disease patients were first documented in patients who followed a supervised aerobic cycling exercise program at moderate intensity, for 45 min/session, three times per week for 8 weeks. The aerobic exercise training resulted in attenuated exercise intolerance compared to baseline [311, 317]. A similar exercise training program resulted in increased peak work capacity, VO_2 peak, cardiac output, and some key mitochondrial enzymes compared to baseline [318]. Favorable training effects were also reported in nine patients who followed a walking or cycling exercise training program including five sessions per week for 32 weeks at duration and intensities similar to the abovementioned studies. VO_2 peak and other variables of exercise capacity were found to be increased with training along with a reduction in serum CK levels [319], indicating that chronic muscle activity may counterbalance muscle wasting and damage. Overall, aerobic exercise interventions are proven to be safe and efficacious for McArdle disease patients [311, 320]. Significant improvement in work capacity without any serious complication for McArdle disease patients were also reported in a similar study, with exercise-related beneficial effects attributed to improved blood flow and mitochondrial metabolism [321].

The effects of resistance exercise have also been evaluated in an adolescent male patient and in seven adult McArdle patients of both sexes [322, 323]. A 15-year-old patient followed a 6-week, light- to moderate-intensity exercise program (two sessions/week, at 65–70% of his one-repetition maximum; 1-RM). After the intervention, his 1-RM power performance improved without any myoglobinuria episodes reported, while, interestingly, he became virtually asymptomatic in terms of exercise limitation [322]. In adult patients, 16-week light to moderate resistance exercise training consisting of two sessions per week, followed by an 8-week detraining period, resulted in a significant beneficial effect on total lean mass, without any major contraindication reported and non-exhibited fixed muscle weakness or limitations in the daily life activities [323].

In conclusion, regular aerobically oriented physical activity or structured exercise programs of moderate intensity are safe and can attenuate

the severity of McArdle disease for these patients [307, 319, 320, 324]. Carbohydrate ingestion prior to exercise is currently the only useful therapy for this disease [313, 314, 320]. This approach appears to improve exercise tolerance to submaximal and maximum workloads and help prevent exercise-induced muscle damage and reduce the threat of renal failure [325, 326]. Some evidence also supports that low to moderate resistance exercises are safe and efficacious for McArdle disease patients. However, HIIT and other forms of high resistance exercises should be avoided.

Conclusions

The paramount importance of the human organism is to maintain metabolic homeostasis. On this basis, ATP levels in skeletal and heart muscle are maintained fairly constant through continuous resynthesis of ATP via anaerobic and aerobic metabolism. The main factor dictating the dominant metabolic pathway and the type of substrate used is exercise intensity, whereas exercise duration, fitness status, gender, diet, and environmental temperature play a secondary role in exercise metabolism. Metabolic pathways do not function independently, but synergistically, by interactions with the exercising muscles and distant organs such as the liver, heart, and brain. Hormones, secreted by cells of the endocrine system, regulate activity of cells in other parts of the body. They are sensitive to exercise-induced stress and modulate metabolism during exercise, not only in skeletal muscle but also in various other organs. Several clinical implications for health benefits of special populations rely on exercise metabolism alterations.

References

1. Phillips SM, Atkinson SA, Tarnopolsky MA, MacDougall JD. Gender differences in leucine kinetics and nitrogen balance in endurance athletes. J Appl Physiol (1985). 1993;75:2134–41.
2. Tarnopolsky MA, Atkinson SA, Phillips SM, MacDougall JD. Carbohydrate loading and metabolism during exercise in men and women. J Appl Physiol (1985). 1995;78:1360–8.
3. Wagenmakers AJ. Muscle amino acid metabolism at rest and during exercise: Role in human physiology and metabolism. Exerc Sport Sci Rev. 1998;26:287–314.
4. Greenhaff PL, Nevill ME, Soderlund K, Bodin K, Boobis LH, Williams C, et al. The metabolic responses of human type i and ii muscle fibres during maximal treadmill sprinting. J Physiol. 1994;478(Pt 1):149–55.
5. Bogdanis GC, Nevill ME, Boobis LH, Lakomy HK, Nevill AM. Recovery of power output and muscle metabolites following 30 s of maximal sprint cycling in man. J Physiol. 1995;482(Pt 2):467–80.
6. Tsintzas OK, Williams C, Boobis L, Greenhaff P. Carbohydrate ingestion and single muscle fiber glycogen metabolism during prolonged running in men. J Appl Physiol (1985). 1996;81:801–9.
7. Sargeant AJ. Structural and functional determinants of human muscle power. Exp Physiol. 2007;92:323–31.
8. Scott W, Stevens J, Binder-Macleod SA. Human skeletal muscle fiber type classifications. Phys Ther. 2001;81:1810–6.
9. Pette D, Staron RS. Myosin isoforms, muscle fiber types, and transitions. Microsc Res Tech. 2000;50:500–9.
10. Schiaffino S, Reggiani C. Fiber types in mammalian skeletal muscles. Physiol Rev. 2011;91:1447–531.
11. Astrand P-O, Rodahl K, Dahl HA, Stromme B. Textbook of work physiology. Physiological bases of exercise. Champaign: Human Kinetics; 2003.
12. Zierath JR, Hawley JA. Skeletal muscle fiber type: Influence on contractile and metabolic properties. PLoS Biol. 2004;2:e348.
13. Sale DG. Influence of exercise and training on motor unit activation. Exerc Sport Sci Rev. 1987;15:95–151.
14. Pette D. The adaptive potential of skeletal muscle fibers. Can J Appl Physiol. 2002;27:423–48.
15. Talbot J, Maves L. Skeletal muscle fiber type: using insights from muscle developmental biology to dissect targets for susceptibility and resistance to muscle disease. Wiley Interdiscip Rev Dev Biol. 2016;5:518–34.
16. Greenhaff PL, Soderlund K, Ren JM, Hultman E. Energy metabolism in single human muscle fibres during intermittent contraction with occluded circulation. J Physiol. 1993;460:443–53.
17. Gray SR, Soderlund K, Ferguson RA. ATP and phosphocreatine utilization in single human muscle fibres during the development of maximal power output at elevated muscle temperatures. J Sports Sci. 2008;26:701–7.
18. Kent-Braun JA, Fitts RH, Christie A. Skeletal muscle fatigue. Compr Physiol. 2012;2:997–1044.
19. Baker JS, McCormick MC, Robergs RA. Interaction among skeletal muscle metabolic energy systems during intense exercise. J Nutr Metab. 2010;2010:905612.

20. Tesch PA, Thorsson A, Fujitsuka N. Creatine phosphate in fiber types of skeletal muscle before and after exhaustive exercise. J Appl Physiol (1985). 1989;66:1756–9.
21. Sahlin K, Soderlund K, Tonkonogi M, Hirakoba K. Phosphocreatine content in single fibers of human muscle after sustained submaximal exercise. Am J Physiol. 1997;273:C172–8.
22. Harris RC, Soderlund K, Hultman E. Elevation of creatine in resting and exercised muscle of normal subjects by creatine supplementation. Clin Sci (Lond). 1992;83:367–74.
23. Solis MY, Artioli GG, Otaduy MCG, Leite CDC, Arruda W, Veiga RR, et al. Effect of age, diet, and tissue type on PCr response to creatine supplementation. J Appl Physiol (1985). 2017;123:407–14.
24. Gollnick PD. Energy metabolism and prolonged exercise. In: Lamb DR, Murray M, editors. Perspectives in exercise science and sports medicine, volume 1:prolonged exercise. Indianapolis: Benchmark Press; 1988. p. 1–42.
25. Gejl KD, Ortenblad N, Andersson E, Plomgaard P, Holmberg HC, Nielsen J. Local depletion of glycogen with supramaximal exercise in human skeletal muscle fibres. J Physiol. 2017;595:2809–21.
26. Hearris MA, Hammond KM, Fell JM, Morton JP. Regulation of muscle glycogen metabolism during exercise: implications for endurance performance and training adaptations. Nutrients. 2018;10:1–21.
27. Gonzalez JT, Fuchs CJ, Betts JA, van Loon LJ. Liver glycogen metabolism during and after prolonged endurance-type exercise. Am J Physiol Endocrinol Metab. 2016;311:E543–53.
28. Hawley JA, Schabort EJ, Noakes TD, Dennis SC. Carbohydrate-loading and exercise performance. An update. Sports Med. 1997;24:73–81.
29. Stevenson EJ, Thelwall PE, Thomas K, Smith F, Brand-Miller J, Trenell MI. Dietary glycemic index influences lipid oxidation but not muscle or liver glycogen oxidation during exercise. Am J Physiol Endocrinol Metab. 2009;296:E1140–7.
30. Gonzalez JT, Fuchs CJ, Smith FE, Thelwall PE, Taylor R, Stevenson EJ, Trenell MI, Cermak NM, van Loon LJ. Ingestion of glucose or sucrose prevents liver but not muscle glycogen depletion during prolonged endurance-type exercise in trained cyclists. Am J Physiol Endocrinol Metab. 2015;309:E1032–9.
31. Tsintzas OK, Williams C, Boobis L, Greenhaff P. Carbohydrate ingestion and glycogen utilization in different muscle fibre types in man. J Physiol. 1995;489(Pt 1):243–50.
32. Tarnopolsky MA, Zawada C, Richmond LB, Carter S, Shearer J, Graham T, et al. Gender differences in carbohydrate loading are related to energy intake. J Appl Physiol (1985). 2001;91:225–30.
33. Walker JL, Heigenhauser GJ, Hultman E, Spriet LL. Dietary carbohydrate, muscle glycogen content, and endurance performance in well-trained women. J Appl Physiol (1985). 2000;88:2151–8.
34. Shin YS. Glycogen storage disease: Clinical, biochemical, and molecular heterogeneity. Semin Pediatr Neurol. 2006;13:115–20.
35. Fleck SJ. Body composition of elite American athletes. Am J Sports Med. 1983;11:398–403.
36. Essen B, Jansson E, Henriksson J, Taylor AW, Saltin B. Metabolic characteristics of fibre types in human skeletal muscle. Acta Physiol Scand. 1975;95:153–65.
37. Watt MJ, Heigenhauser GJ, Spriet LL. Intramuscular triacylglycerol utilization in human skeletal muscle during exercise: Is there a controversy? J Appl Physiol (1985). 2002;93:1185–95.
38. Wendling PS, Peters SJ, Heigenhauser GJ, Spriet LL. Variability of triacylglycerol content in human skeletal muscle biopsy samples. J Appl Physiol (1985). 1996;81:1150–5.
39. Stellingwerff T, Boon H, Jonkers RA, Senden JM, Spriet LL, Koopman R, et al. Significant intramyocellular lipid use during prolonged cycling in endurance-trained males as assessed by three different methodologies. Am J Physiol Endocrinol Metab. 2007;292:E1715–23.
40. Newsholme EA, Leech AR. Biochemistry for the medical sciences. Chichester: Wiley; 1983.
41. Metzeger JM. Mechanism of chemomechanical coupling in skeletal muscle during work. In: Lamb DR, Gisolfi CV, editors. Energy metabolism in exercise and sport, vol. 5. Dubuque: Brown & Benchmark; 1992. p. 1–51.
42. Spriet LL. Anaerobic metabolism during exercise. In: Hargreaves M, Spriet L, editors. Exercise metabolism. Champaign: Human Kinetics; 2006. p. 7–27.
43. Kenney LW, Wilmore JH, Costill DL. Physiology of sport and exercise. 5th ed. Champaign: Human Kinetics, Champaign, IL; 2012.
44. MacLaren D, Morton J. Biochemistry for sport and exercise metabolism. Champaign: Wiley-Blackwell; 2011.
45. Sahlin K. Metabolic factors in fatigue. In: Hargreaves M, Spriet L, editors. Exercise metabolism. Champaign: Human Kinetics; 2006. p. 163–86.
46. Berg JM, Tymoczko JL, Stryer L. Biochemistry. 5th ed. New York: W.H. Freeman and Company; 2002.
47. Chasiotis D. The regulation of glycogen phosphorylase and glycogen breakdown in human skeletal muscle. Acta Physiol Scand Suppl. 1983;518:1–68.
48. Rush JW, Spriet LL. Skeletal muscle glycogen phosphorylase a kinetics: Effects of adenine nucleotides and caffeine. J Appl Physiol (1985). 2001;91:2071–8.
49. Costill DL, Coyle E, Dalsky G, Evans W, Fink W, Hoopes D. Effects of elevated plasma FFA and insulin on muscle glycogen usage during exercise. J Appl Physiol Respir Environ Exerc Physiol. 1977;43:695–9.
50. Dyck DJ, Peters SJ, Wendling PS, Chesley A, Hultman E, Spriet LL. Regulation of muscle glycogen phosphorylase activity during intense aerobic cycling with elevated FFA. Am J Phys. 1996;270:E116–25.

51. Hargreaves M, McConell G, Proietto J. Influence of muscle glycogen on glycogenolysis and glucose uptake during exercise in humans. J Appl Physiol (1985). 1995;78:288–92.
52. Howlett KF, Spriet LL, Hargreaves M. Carbohydrate metabolism during exercise in females: effect of reduced fat availability. Metabolism. 2001;50:481–7.
53. Ferguson BS, Rogatzki MJ, Goodwin ML, Kane DA, Rightmire Z, Gladden LB. Lactate metabolism: historical context, prior misinterpretations, and current understanding. Eur J Appl Physiol. 2018;118:691–728.
54. Brooks GA. Lactate: link between glycolytic and oxidative metabolism. Sports Med. 2007;37:341–3.
55. O'Brien MJ, Viguie CA, Mazzeo RS, Brooks GA. Carbohydrate dependence during marathon running. Med Sci Sports Exerc. 1993;25:1009–17.
56. Robergs RA, Roberts SO. Exercise physiology. Exercise, performance, and clinical applications. Boston: WCB, McGraw-Hill; 1997.
57. Gladden LB. Lactate metabolism: a new paradigm for the third millennium. J Physiol. 2004;558:5–30.
58. Margaria R, Edwards RHT, Dill DB. The possible mechanisms of contracting and paying the oxygen debt and the role of lactic acid in muscular contraction. Am J Physiol. 1933;106:689–715.
59. Bergman BC, Wolfel EE, Butterfield GE, Lopaschuk GD, Casazza GA, Horning MA, et al. Active muscle and whole body lactate kinetics after endurance training in men. J Appl Physiol (1985). 1999;87:1684–96.
60. Nielsen HB, Clemmesen JO, Skak C, Ott P, Secher NH. Attenuated hepatosplanchnic uptake of lactate during intense exercise in humans. J Appl Physiol (1985). 2002;92:1677–83.
61. Mangia S, Garreffa G, Bianciardi M, Giove F, Di Salle F, Maraviglia B. The aerobic brain: lactate decrease at the onset of neural activity. Neuroscience. 2003;118:7–10.
62. Green JM, Hornsby JH, Pritchett RC, Pritchett K. Lactate threshold comparison in anaerobic vs. aerobic athletes and untrained participants. Int J Exerc Sci. 2014;7:329–38.
63. Brooks GA. The lactate shuttle during exercise and recovery. Med Sci Sports Exerc. 1986;18:360–8.
64. Goodwin ML, Gladden LB, Nijsten MW, Jones KB. Lactate and cancer: revisiting the warburg effect in an era of lactate shuttling. Front Nutr. 2014;1:27.
65. Nijsten MW, van Dam GM. Hypothesis: using the warburg effect against cancer by reducing glucose and providing lactate. Med Hypotheses. 2009;73:48–51.
66. Wasserman K, McIlroy MB. Detecting the threshold of anaerobic metabolism in cardiac patients during exercise. Am J Cardiol. 1964;14:844–52.
67. Yoshida T, Nagata A, Muro M, Takeuchi N, Suda Y. The validity of anaerobic threshold determination by a douglas bag method compared with arterial blood lactate concentration. Eur J Appl Physiol Occup Physiol. 1981;46:423–30.
68. Sjodin B, Jacobs I, Svedenhag J. Changes in onset of blood lactate accumulation (obla) and muscle enzymes after training at obla. Eur J Appl Physiol Occup Physiol. 1982;49:45–57.
69. Coyle EF. Integration of the physiological factors determining endurance performance ability. Exerc Sport Sci Rev. 1995;23:25–63.
70. Ghosh AK. Anaerobic threshold: its concept and role in endurance sport. Malays J Med Sci. 2004;11:24–36.
71. Agostoni P, Corra U, Cattadori G, Veglia F, Battaia E, La Gioia R, et al. Prognostic value of indeterminable anaerobic threshold in heart failure. Circ Heart Fail. 2013;6:977–87.
72. Rusko H, Rahkila P, Karvinen E. Anaerobic threshold, skeletal muscle enzymes and fiber composition in young female cross-country skiers. Acta Physiol Scand. 1980;108:263–8.
73. Bassett DR Jr, Howley ET. Limiting factors for maximum oxygen uptake and determinants of endurance performance. Med Sci Sports Exerc. 2000;32:70–84.
74. Kenney LWW, Costill JH, D.L. Physiology of sport and exercise. 5th ed. Champaign: Human Kinetics; 2012.
75. Jeukendrup AE, Gleeson M. Sports nutrition. An introduction to energy production and performance. 1st ed. Champaign: Human Kinetics; 2004.
76. Trefts E, Williams AS, Wasserman DH. Exercise and the regulation of hepatic metabolism. Prog Mol Biol Transl Sci. 2015;135:203–25.
77. Kjaer M. Hepatic metabolism during exercise. In: Hargreaves M, Spriet L, editors. Exercise metabolism. Champaign: Human Kinetics; 2006. p. 45–70.
78. Lavoie C, Ducros F, Bourque J, Langelier H, Chiasson JL. Glucose metabolism during exercise in man: the role of insulin and glucagon in the regulation of hepatic glucose production and gluconeogenesis. Can J Physiol Pharmacol. 1997;75:26–35.
79. Tuttle KR, Marker JC, Dalsky GP, Schwartz NS, Shah SD, Clutter WE, et al. Glucagon, not insulin, may play a secondary role in defense against hypoglycemia during exercise. Am J Physiol. 1988;254:E713–9.
80. Rose AJ, Richter EA. Skeletal muscle glucose uptake during exercise: how is it regulated? Physiology (Bethesda). 2005;20:260–70.
81. Ahlborg G, Felig P, Hagenfeldt L, Hendler R, Wahren J. Substrate turnover during prolonged exercise in man. Splanchnic and leg metabolism of glucose, free fatty acids, and amino acids. J Clin Invest. 1974;53:1080–90.
82. Ahlborg G, Felig P. Lactate and glucose exchange across the forearm, legs, and splanchnic bed during and after prolonged leg exercise. J Clin Invest. 1982;69:45–54.
83. Katz A, Broberg S, Sahlin K, Wahren J. Leg glucose uptake during maximal dynamic exercise in humans. Am J Physiol. 1986;251:E65–70.
84. Jeukendrup AE, Raben A, Gijsen A, Stegen JH, Brouns F, Saris WH, et al. Glucose kinetics during

prolonged exercise in highly trained human subjects: effect of glucose ingestion. J Physiol. 1999;515(Pt 2):579–89.
85. Angus DJ, Febbraio MA, Hargreaves M. Plasma glucose kinetics during prolonged exercise in trained humans when fed carbohydrate. Am J Physiol Endocrinol Metab. 2002;283:E573–7.
86. Richter EA, Hargreaves M. Exercise, GLUT4, and skeletal muscle glucose uptake. Physiol Rev. 2013;93:993–1017.
87. Merry TL, McConell GK. Skeletal muscle glucose uptake during exercise: a focus on reactive oxygen species and nitric oxide signaling. IUBMB Life. 2009;61:479–84.
88. Richter EA, Ruderman NB. AMPK and the biochemistry of exercise: implications for human health and disease. Biochem J. 2009;418:261–75.
89. Katz A. Role of reactive oxygen species in regulation of glucose transport in skeletal muscle during exercise. J Physiol. 2016;594:2787–94.
90. Watt MJ, Howlett KF, Febbraio MA, Spriet LL, Hargreaves M. Adrenaline increases skeletal muscle glycogenolysis, pyruvate dehydrogenase activation and carbohydrate oxidation during moderate exercise in humans. J Physiol. 2001;534:269–78.
91. Steensberg A, van Hall G, Keller C, Osada T, Schjerling P, Pedersen BK, et al. Muscle glycogen content and glucose uptake during exercise in humans: Influence of prior exercise and dietary manipulation. J Physiol. 2002;541:273–81.
92. Pedersen BK, Akerstrom TC, Nielsen AR, Fischer CP. Role of myokines in exercise and metabolism. J Appl Physiol (1985). 2007;103:1093–8.
93. Pedersen L, Hojman P. Muscle-to-organ cross talk mediated by myokines. Adipocyte. 2012;1:164–7.
94. Pedersen BK, Febbraio MA. Muscle as an endocrine organ: focus on muscle-derived interleukin-6. Physiol Rev. 2008;88:1379–406.
95. Febbraio MA, Hiscock N, Sacchetti M, Fischer CP, Pedersen BK. Interleukin-6 is a novel factor mediating glucose homeostasis during skeletal muscle contraction. Diabetes. 2004;53:1643–8.
96. Pedersen L, Pilegaard H, Hansen J, Brandt C, Adser H, Hidalgo J, et al. Exercise-induced liver chemokine CXCL-1 expression is linked to muscle-derived interleukin-6 expression. J Physiol. 2011;589:1409–20.
97. Pedersen BK, Febbraio M. Muscle-derived interleukin-6--a possible link between skeletal muscle, adipose tissue, liver, and brain. Brain Behav Immun. 2005;19:371–6.
98. Spriet LL. Regulation of skeletal muscle fat oxidation during exercise in humans. Med Sci Sports Exerc. 2002;34:1477–84.
99. Horowitz JF. Adipose tissue lipid mobilization during exercise. In: Hargreaves M, Spriet L, editors. Exercise metabolism. Champaign: Human Kinetics; 2006. p. 89–104.
100. Romijn JA, Coyle EF, Sidossis LS, Gastaldelli A, Horowitz JF, Endert E, et al. Regulation of endogenous fat and carbohydrate metabolism in relation to exercise intensity and duration. Am J Physiol. 1993;265:E380–91.
101. Romijn JA, Coyle EF, Sidossis LS, Zhang XJ, Wolfe RR. Relationship between fatty acid delivery and fatty acid oxidation during strenuous exercise. J Appl Physiol (1985). 1995;79:1939–45.
102. Luiken JJ, Glatz JF, Bonen A. Fatty acid transport proteins facilitate fatty acid uptake in skeletal muscle. Can J Appl Physiol. 2000;25:333–52.
103. Hargreaves M, Spriet LL. Exercise metabolism: fuels for the fire. Cold Spring Harb Perspect Med. 2018;8(8):1–15.
104. Holloway GP, Luiken JJ, Glatz JF, Spriet LL, Bonen A. Contribution of FAT/CD36 to the regulation of skeletal muscle fatty acid oxidation: an overview. Acta Physiol (Oxf). 2008;194:293–309.
105. Holloway GP, Bonen A, Spriet LL. Regulation of skeletal muscle mitochondrial fatty acid metabolism in lean and obese individuals. Am J Clin Nutr. 2009;89:455S–62S.
106. Tarnopolsky MA. Protein metabolism in strength and endurance activities. In: Lamb DR, Murray R, editors. The metabolic bases of performance in exercise and sport. Carmel: Cooper Publishing Group; 1999. p. 125–63.
107. Gibala MJ. Effect of exercise on skeletal muscle protein and amino acid metabolism in humans. In: Hargreaves M, Spriet L, editors. Exercise metabolism. Champaign: Human Kinetics; 2006. p. 137–61.
108. Wagenmakers AJ, Brookes JH, Coakley JH, Reilly T, Edwards RH. Exercise-induced activation of the branched-chain 2-oxo acid dehydrogenase in human muscle. Eur J Appl Physiol Occup Physiol. 1989;59:159–67.
109. Rush JW, MacLean DA, Hultman E, Graham TE. Exercise causes branched-chain oxoacid dehydrogenase dephosphorylation but not AMP deaminase binding. J Appl Physiol (1985). 1995;78:2193–200.
110. van Hall G, MacLean DA, Saltin B, Wagenmakers AJ. Mechanisms of activation of muscle branched-chain alpha-keto acid dehydrogenase during exercise in man. J Physiol. 1996;494(Pt 3):899–905.
111. Wagenmakers AJ, Beckers EJ, Brouns F, Kuipers H, Soeters PB, van der Vusse GJ, et al. Carbohydrate supplementation, glycogen depletion, and amino acid metabolism during exercise. Am J Physiol. 1991;260:E883–90.
112. Lemon PW, Mullin JP. Effect of initial muscle glycogen levels on protein catabolism during exercise. J Appl Physiol Respir Environ Exerc Physiol. 1980;48:624–9.
113. Gibala MJ. Regulation of skeletal muscle amino acid metabolism during exercise. Int J Sport Nutr Exerc Metab. 2001;11:87–108.
114. Phillips SM, Tipton KD, Aarsland A, Wolf SE, Wolfe RR. Mixed muscle protein synthesis and breakdown after resistance exercise in humans. Am J Physiol. 1997;273:E99–107.

115. Romijn JA, Coyle EF, Sidossis LS, Rosenblatt J, Wolfe RR. Substrate metabolism during different exercise intensities in endurance-trained women. J Appl Physiol (1985). 2000;88:1707–14.
116. Achten J, Gleeson M, Jeukendrup AE. Determination of the exercise intensity that elicits maximal fat oxidation. Med Sci Sports Exerc. 2002;34:92–7.
117. Coyle EF, Jeukendrup AE, Wagenmakers AJ, Saris WH. Fatty acid oxidation is directly regulated by carbohydrate metabolism during exercise. Am J Physiol. 1997;273:E268–75.
118. Sidossis LS, Gastaldelli A, Klein S, Wolfe RR. Regulation of plasma fatty acid oxidation during low- and high-intensity exercise. Am J Physiol. 1997;272:E1065–70.
119. Bergman BC, Brooks GA. Respiratory gas-exchange ratios during graded exercise in fed and fasted trained and untrained men. J Appl Physiol (1985). 1999;86:479–87.
120. Edwards HT, Margaria R, Dill DB. Metabolic rate, blood sugar and utilization of carbohydrate. Am J Physiol. 1934;108:203–9.
121. Martin WH. Effect of endurance training on fatty acid metabolism during whole body exercise. Med Sci Sports Exerc. 1997;29:635–9.
122. Kiens B. Effect of endurance training on fatty acid metabolism: Local adaptations. Med Sci Sports Exerc. 1997;29:640–5.
123. Nicklas BJ. Effects of endurance exercise on adipose tissue metabolism. Exerc Sport Sci Rev. 1997;25:77–103.
124. Krogh AL, J. The relative value of fat and carbohydrate as sources of muscular energy. Biochem J. 1920;14:290–363.
125. Jeukendrup AE. Periodized nutrition for athletes. Sports Med. 2017;47:51–63.
126. Wright DA, Sherman WM, Dernbach AR. Carbohydrate feedings before, during, or in combination improve cycling endurance performance. J Appl Physiol (1985). 1991;71:1082–8.
127. Chryssanthopoulos C, Williams C. Pre-exercise carbohydrate meal and endurance running capacity when carbohydrates are ingested during exercise. Int J Sports Med. 1997;18:543–8.
128. Chryssanthopoulos C, Williams C, Nowitz A, Kotsiopoulou C, Vleck V. The effect of a high carbohydrate meal on endurance running capacity. Int J Sport Nutr Exerc Metab. 2002;12:157–71.
129. Montain SJ, Hopper MK, Coggan AR, Coyle EF. Exercise metabolism at different time intervals after a meal. J Appl Physiol (1985). 1991;70:882–8.
130. Febbraio MA. Temperature, muscle metabolism and performance. In: Lamb DR, Murray R, editors. The metabolic basis of performance in exercise and sport. Traverse City: Cooper Publishing Group; 1999. p. 315–53.
131. Devries MC. Sex-based differences in endurance exercise muscle metabolism: impact on exercise and nutritional strategies to optimize health and performance in women. Exp Physiol. 2016;101:243–9.
132. Brockman NK, Yardley JE. Sex-related differences in fuel utilization and hormonal response to exercise: implications for individuals with type 1 diabetes. Appl Physiol Nutr Metab. 2018;43(6):541–52.
133. Tarnopolsky LJ, MacDougall JD, Atkinson SA, Tarnopolsky MA, Sutton JR. Gender differences in substrate for endurance exercise. J Appl Physiol (1985). 1990;68:302–8.
134. Horton TJ, Pagliassotti MJ, Hobbs K, Hill JO. Fuel metabolism in men and women during and after long-duration exercise. J Appl Physiol (1985). 1998;85:1823–32.
135. Marliss EB, Kreisman SH, Manzon A, Halter JB, Vranic M, Nessim SJ. Gender differences in glucoregulatory responses to intense exercise. J Appl Physiol (1985). 2000;88:457–66.
136. Hamadeh MJ, Devries MC, Tarnopolsky MA. Estrogen supplementation reduces whole body leucine and carbohydrate oxidation and increases lipid oxidation in men during endurance exercise. J Clin Endocrinol Metab. 2005;90:3592–9.
137. Boisseau N, Delamarche P. Metabolic and hormonal responses to exercise in children and adolescents. Sports Med. 2000;30:405–22.
138. Oosthuyse T, Bosch AN. The effect of the menstrual cycle on exercise metabolism: implications for exercise performance in eumenorrhoeic women. Sports Med. 2010;40:207–27.
139. Numao S, Hayashi Y, Katayama Y, Matsuo T, Tanaka K. Sex differences in substrate oxidation during aerobic exercise in obese men and postmenopausal obese women. Metabolism. 2009;58:1312–9.
140. Devries MC, Hamadeh MJ, Graham TE, Tarnopolsky MA. 17beta-estradiol supplementation decreases glucose rate of appearance and disappearance with no effect on glycogen utilization during moderate intensity exercise in men. J Clin Endocrinol Metab. 2005;90:6218–25.
141. Devries MC, Hamadeh MJ, Phillips SM, Tarnopolsky MA. Menstrual cycle phase and sex influence muscle glycogen utilization and glucose turnover during moderate-intensity endurance exercise. Am J Physiol Regul Integr Comp Physiol. 2006;291:R1120–8.
142. Isacco L, Duche P, Boisseau N. Influence of hormonal status on substrate utilization at rest and during exercise in the female population. Sports Med. 2012;42:327–42.
143. Kraemer RR, Heleniak RJ, Tryniecki JL, Kraemer GR, Okazaki NJ, Castracane VD. Follicular and luteal phase hormonal responses to low-volume resistive exercise. Med Sci Sports Exerc. 1995;27:809–17.
144. Riddell MC, Partington SL, Stupka N, Armstrong D, Rennie C, Tarnopolsky MA. Substrate utilization during exercise performed with and without glucose ingestion in female and male endurance trained athletes. Int J Sport Nutr Exerc Metab. 2003;13:407–21.
145. Copeland JL, Consitt LA, Tremblay MS. Hormonal responses to endurance and resistance exercise in females aged 19-69 years. J Gerontol A Biol Sci Med Sci. 2002;57:B158–65.

146. Fragala MS, Kraemer WJ, Denegar CR, Maresh CM, Mastro AM, Volek JS. Neuroendocrine-immune interactions and responses to exercise. Sports Med. 2011;41:621–39.
147. Roepstorff C, Steffensen CH, Madsen M, Stallknecht B, Kanstrup IL, Richter EA, et al. Gender differences in substrate utilization during submaximal exercise in endurance-trained subjects. Am J Physiol Endocrinol Metab. 2002;282:E435–47.
148. Steffensen CH, Roepstorff C, Madsen M, Kiens B. Myocellular triacylglycerol breakdown in females but not in males during exercise. Am J Physiol Endocrinol Metab. 2002;282:E634–42.
149. Taegtmeyer H. Cardiomyocyte metabolism: all is in flux. Muscle Fundamental Biol Mech Dis. 2012;1:187–202.
150. Abdel-aleem S, Lowe JE, editors. Cardiac metabolism in health and disease. Dordrecht: Springer Science+Business Media, B.V; 1998.
151. Stanley WC, Recchia FA, Lopaschuk GD. Myocardial substrate metabolism in the normal and failing heart. Physiol Rev. 2005;85:1093–129.
152. Lopaschuk GD, Dhalla NS, editors. Cardiac energy metabolism in health and disease. New York: Springer; 2014.
153. Schwarzer M, Doenst T, editors. The scientist's guide to cardiac metabolism. Amsterdam: Academic; 2016.
154. Taegtmeyer H, Young ME, Lopaschuk GD, Abel ED, Brunengraber H, Darley-Usmar V, et al. American Heart Association Council on Basic Cardiovascular Sciences. Assessing cardiac metabolism: a scientific statement from the American heart association. Circ Res. 2016;118:1659–701.
155. Lopaschuk GD, Kelly DP. Signalling in cardiac metabolism. Cardiovasc Res. 2008;79:205–7.
156. Keul J. Myocardial metabolism in athletes. In: Pernow B, Saltin B, editors. Muscle metabolism during exercise. New York/London: Plenum Press; 1971.
157. Kaijser L, Lassers BW, Wahlqvist ML, Carlson LA. Myocardial lipid and carbohydrate metabolism in fasting men during prolonged exercise. J Appl Physiol. 1972;32:847–58.
158. Lassers BW, Wahlqvist ML, Kaijser L, Carlson LA. Effect of nicotinic acid on myocardial metabolism in man at rest and during exercise. J Appl Physiol. 1972;33:72–80.
159. Gertz EW, Wisneski JA, Stanley WC, Neese RA. Myocardial substrate utilization during exercise in humans. Dual carbon-labeled carbohydrate isotope experiments. J Clin Invest. 1988;82:2017–25.
160. Stanley WC. Myocardial lactate metabolism during exercise. Med Sci Sports Exerc. 1991;23:920–4.
161. Kaijser L, Berglund B. Myocardial lactate extraction and release at rest and during heavy exercise in healthy men. Acta Physiol Scand. 1992;144:39–45.
162. Kemppainen J, Fujimoto T, Kalliokoski KK, Viljanen T, Nuutila P, Knuuti J. Myocardial and skeletal muscle glucose uptake during exercise in humans. J Physiol. 2002;542:403–12.
163. Bing RJ. The metabolism of the heart. Trans Am Coll Cardiol. 1955;5:8–14.
164. Bremer J, Wojtczak AB. Factors controlling the rate of fatty acid -oxidation in rat liver mitochondria. Biochim Biophys Acta. 1972;280:515–30.
165. Williamson JR, Krebs HA. Acetoacetate as fuel of respiration in the perfused rat heart. Biochem J. 1961;80:540–7.
166. Zimmerman AN, Meijler FL, Hulsmann WC. The inhibitory effect of acetoacetate on myocardial contraction. Lancet. 1962;2:757–8.
167. Taegtmeyer H, Hems R, Krebs HA. Utilization of energy-providing substrates in the isolated working rat heart. Biochem J. 1980;186:701–11.
168. Krebs HA. Some aspects of the regulation of fuel supply in omnivorous animals. Adv Enzyme Regul. 1972;10:397–420.
169. Balaban RS. Cardiac energy metabolism homeostasis: Role of cytosolic calcium. J Mol Cell Cardiol. 2002;34:1259–71.
170. Balaban RS, Kantor HL, Katz LA, Briggs RW. Relation between work and phosphate metabolite in the in vivo paced mammalian heart. Science. 1986;232:1121–3.
171. Kupriyanov VV, Lakomkin VL, Kapelko VI, Steinschneider A, Ruuge EK, Saks VA. Dissociation of adenosine triphosphate levels and contractile function in isovolumic hearts perfused with 2-deoxyglucose. J Mol Cell Cardiol. 1987;19:729–40.
172. Stanley WC, Chandler MP. Energy metabolism in the normal and failing heart: potential for therapeutic interventions. Heart Fail Rev. 2002;7:115–30.
173. Gollnick PD. Metabolic regulation in skeletal muscle: influence of endurance training as exerted by mitochondrial protein concentration. Acta Physiol Scand Suppl. 1986;556:53–66.
174. Kuno S, Ogawa T, Katsuta S, Itai Y. In vivo human myocardial metabolism during aerobic exercise by phosphorus-31 nuclear magnetic resonance spectroscopy. Eur J Appl Physiol Occup Physiol. 1994;69:488–91.
175. Balaban RS, Heineman FW. Control of mitochondrial respiration in the heart in vivo. Mol Cell Biochem. 1989;89:191–7.
176. Grubbstrom J, Berglund B, Kaijser L. Myocardial blood flow and lactate metabolism at rest and during exercise with reduced arterial oxygen content. Acta Physiol Scand. 1991;142:467–74.
177. Jansson E, Sylven C. Activities of key enzymes in the energy metabolism of human myocardial and skeletal muscle. Clin Physiol. 1986;6:465–71.
178. Conway MA, Bristow JD, Blackledge MJ, Rajagopalan B, Radda GK. Cardiac metabolism during exercise in healthy volunteers measured by 31P magnetic resonance spectroscopy. Br Heart J. 1991;65:25–30.
179. Arany Z, He H, Lin J, Hoyer K, Handschin C, Toka O, et al. Transcriptional coactivator pgc-1 alpha controls the energy state and contractile function of cardiac muscle. Cell Metab. 2005;1:259–71.

180. Goodwin GW, Taegtmeyer H. Improved energy homeostasis of the heart in the metabolic state of exercise. Am J Physiol Heart Circ Physiol. 2000;279:H1490–501.
181. Goodwin GW, Taylor CS, Taegtmeyer H. Regulation of energy metabolism of the heart during acute increase in heart work. J Biol Chem. 1998;273:29530–9.
182. Goodwin GW, Ahmad F, Doenst T, Taegtmeyer H. Energy provision from glycogen, glucose, and fatty acids on adrenergic stimulation of isolated working rat hearts. Am J Physiol. 1998;274:H1239–47.
183. Chatham JC. Lactate – the forgotten fuel! J Physiol. 2002;542:333.
184. Lewis GD, Farrell L, Wood MJ, Martinovic M, Arany Z, Rowe GC, et al. Metabolic signatures of exercise in human plasma. Sci Transl Med. 2010;542:33ra37.
185. Mann N, Rosenzweig A. Can exercise teach us how to treat heart disease? Circulation. 2012;126:2625–35.
186. Finkel T, Deng CX, Mostoslavsky R. Recent progress in the biology and physiology of sirtuins. Nature. 2009;460:587–91.
187. Ferrara N, Rinaldi B, Corbi G, Conti V, Stiuso P, Boccuti S, et al. Exercise training promotes SIRT1 activity in aged rats. Rejuvenation Res. 2008;11:139–50.
188. Pillai VB, Sundaresan NR, Jeevanandam V, Gupta MP. Mitochondrial SIRT3 and heart disease. Cardiovasc Res. 2010;88:250–6.
189. Sundaresan NR, Gupta M, Kim G, Rajamohan SB, Isbatan A, Gupta MP. SIRT3 blocks the cardiac hypertrophic response by augmenting Foxo3a-dependent antioxidant defense mechanisms in mice. J Clin Invest. 2009;119:2758–71.
190. Hafner AV, Dai J, Gomes AP, Xiao CY, Palmeira CM, Rosenzweig A, et al. Regulation of the mPTP by SIRT3-mediated deacetylation of CypD at lysine 166 suppresses age-related cardiac hypertrophy. Aging (Albany NY). 2010;2:914–23.
191. Arany Z, Foo SY, Ma Y, Ruas JL, Bommi-Reddy A, Girnun G, et al. HIF-independent regulation of VEGF and angiogenesis by the transcriptional coactivator PGC-1alpha. Nature. 2008;451:1008–12.
192. Young ME, McNulty P, Taegtmeyer H. Adaptation and maladaptation of the heart in diabetes: Part ii: potential mechanisms. Circulation. 2002;105:1861–70.
193. Hellsten Y, Nyberg M. Cardiovascular adaptations to exercise training. Compr Physiol. 2015;6:1–32.
194. McKenzie DC. Respiratory physiology: adaptations to high-level exercise. Br J Sports Med. 2012;46:381–4.
195. Gabriel DA, Kamen G, Frost G. Neural adaptations to resistive exercise: mechanisms and recommendations for training practices. Sports Med. 2006;36:133–49.
196. Hedayatpour N, Falla D. Physiological and neural adaptations to eccentric exercise: mechanisms and considerations for training. Biomed Res Int. 2015;2015:193741.
197. Holloszy JO, Coyle EF. Adaptations of skeletal muscle to endurance exercise and their metabolic consequences. J Appl Physiol Respir Environ Exerc Physiol. 1984;56:831–8.
198. Lundby C, Jacobs RA. Adaptations of skeletal muscle mitochondria to exercise training. Exp Physiol. 2016;101:17–22.
199. Hood DA, Uguccioni G, Vainshtein A, D'Souza D. Mechanisms of exercise-induced mitochondrial biogenesis in skeletal muscle: implications for health and disease. Compr Physiol. 2011;1:1119–34.
200. Kiens B, Essen-Gustavsson B, Christensen NJ, Saltin B. Skeletal muscle substrate utilization during submaximal exercise in man: effect of endurance training. J Physiol. 1993;469:459–78.
201. Laughlin MH, Roseguini B. Mechanisms for exercise training-induced increases in skeletal muscle blood flow capacity: differences with interval sprint training versus aerobic endurance training. J Physiol Pharmacol. 2008;59(Suppl 7):71–88.
202. Green HJ, Jones S, Ball-Burnett ME, Smith D, Livesey J, Farrance BW. Early muscular and metabolic adaptations to prolonged exercise training in humans. J Appl Physiol (1985). 1991;70:2032–8.
203. Chesley A, Heigenhauser GJ, Spriet LL. Regulation of muscle glycogen phosphorylase activity following short-term endurance training. Am J Physiol. 1996;270:E328–35.
204. Phillips SM, Green HJ, Tarnopolsky MA, Heigenhauser GJ, Grant SM. Progressive effect of endurance training on metabolic adaptations in working skeletal muscle. Am J Physiol. 1996;270:E265–72.
205. Coggan AR, Swanson SC, Mendenhall LA, Habash DL, Kien CL. Effect of endurance training on hepatic glycogenolysis and gluconeogenesis during prolonged exercise in men. Am J Physiol. 1995;268:E375–83.
206. Mendenhall LA, Swanson SC, Habash DL, Coggan AR. Ten days of exercise training reduces glucose production and utilization during moderate-intensity exercise. Am J Physiol. 1994;266:E136–43.
207. Bergman BC, Horning MA, Casazza GA, Wolfel EE, Butterfield GE, Brooks GA. Endurance training increases gluconeogenesis during rest and exercise in men. Am J Physiol Endocrinol Metab. 2000;278:E244–51.
208. Coggan AR, Kohrt WM, Spina RJ, Bier DM, Holloszy JO. Endurance training decreases plasma glucose turnover and oxidation during moderate-intensity exercise in men. J Appl Physiol (1985). 1990;68:990–6.
209. Phillips SM, Green HJ, Tarnopolsky MA, Heigenhauser GF, Hill RE, Grant SM. Effects of training duration on substrate turnover and oxidation during exercise. J Appl Physiol (1985). 1996;81:2182–91.
210. Kristiansen S, Gade J, Wojtaszewski JF, Kiens B, Richter EA. Glucose uptake is increased in trained

vs. Untrained muscle during heavy exercise. J Appl Physiol (1985). 2000;89:1151–8.
211. Phillips SM. Endurance training-induced adaptations in substrate turnover and oxidation. In: Hargreaves M, Spriet L, editors. Exercise metabolism. Champaign: Human Kinetics; 2006. p. 187–213.
212. Purdom T, Kravitz L, Dokladny K, Mermier C. Understanding the factors that effect maximal fat oxidation. J Int Soc Sports Nutr. 2018;15:3.
213. MacRae HS, Dennis SC, Bosch AN, Noakes TD. Effects of training on lactate production and removal during progressive exercise in humans. J Appl Physiol (1985). 1992;72:1649–56.
214. Messonnier LA, Emhoff CA, Fattor JA, Horning MA, Carlson TJ, Brooks GA. Lactate kinetics at the lactate threshold in trained and untrained men. J Appl Physiol (1985). 2013;114:1593–602.
215. Dubouchaud H, Butterfield GE, Wolfel EE, Bergman BC, Brooks GA. Endurance training, expression, and physiology of LDH, MCT1, and MCT4 in human skeletal muscle. Am J Physiol Endocrinol Metab. 2000;278:E571–9.
216. Philippou A, Maridaki M, Tenta R, Koutsilieris M. Hormonal responses following eccentric exercise in humans. Hormones (Athens). 2017;16:405–13.
217. Weicker H, Strobel G. Endocrine regulation of metabolism during exercise. In: Steinacker JM, Ward SA, editors. The physiology and pathophysiology of exercise tolerance. New York: Springer Science & Business Media; 1996.
218. Benardot D, editor. Advanced sports nutrition. 2nd ed. Champaign: Human Kinetics; 2012.
219. Nesher R, Karl IE, Kipnis DM. Dissociation of effects of insulin and contraction on glucose transport in rat epitrochlearis muscle. Am J Physiol. 1985;249:C226–32.
220. Etgen GJ Jr, Memon AR, Thompson GA Jr, Ivy JL. Insulin- and contraction-stimulated translocation of GTP-binding proteins and GLUT4 protein in skeletal muscle. J Biol Chem. 1993;268:20164–9.
221. Farrell PA. Exercise effects on regulation of energy metabolism by pancreatic and gut hormones. Perspectives in Exerc Science and Sports Med Energy Supply in Exercise and Sport. 1992;5:383–434.
222. Ploug T, Galbo H, Richter EA. Increased muscle glucose uptake during contractions: no need for insulin. Am J Physiol. 1984;247:E726–31.
223. Ploug T, Galbo H, Ohkuwa T, Tranum-Jensen J, Vinten J. Kinetics of glucose transport in rat skeletal muscle membrane vesicles: effects of insulin and contractions. Am J Physiol. 1992;262:E700–11.
224. Hamada T, Arias EB, Cartee GD. Increased submaximal insulin-stimulated glucose uptake in mouse skeletal muscle after treadmill exercise. J Appl Physiol (1985). 2006;101:1368–76.
225. Richter EA, Ploug T, Galbo H. Increased muscle glucose uptake after exercise. No need for insulin during exercise. Diabetes. 1985;34:1041–8.
226. Slentz CA, Gulve EA, Rodnick KJ, Henriksen EJ, Youn JH, Holloszy JO. Glucose transporters and maximal transport are increased in endurance-trained rat soleus. J Appl Physiol (1985). 1992;73:486–92.
227. Flores-Riveros JR, Kaestner KH, Thompson KS, Lane MD. Cyclic AMP-induced transcriptional repression of the insulin-responsive glucose transporter (GLUT4) gene: identification of a promoter region required for down-regulation of transcription. Biochem Biophys Res Commun. 1993;194:1148–54.
228. Goodyear LJ, Hirshman MF, Valyou PM, Horton ES. Glucose transporter number, function, and subcellular distribution in rat skeletal muscle after exercise training. Diabetes. 1992;41:1091–9.
229. Neufer PD, Shinebarger MH, Dohm GL. Effect of training and detraining on skeletal muscle glucose transporter (GLUT4) content in rats. Can J Physiol Pharmacol. 1992;70:1286–90.
230. Kono T, Robinson FW, Blevins TL, Ezaki O. Evidence that translocation of the glucose transport activity is the major mechanism of insulin action on glucose transport in fat cells. J Biol Chem. 1982;257:10942–7.
231. Goodyear LJ, Kahn BB. Exercise, glucose transport, and insulin sensitivity. Annu Rev Med. 1998;49:235–61.
232. Wallberg-Henriksson H. Exercise and diabetes mellitus. Exerc Sport Sci Rev. 1992;20:339–68.
233. Richter EA, Galbo H, Sonne B, Holst JJ, Christensen NJ. Adrenal medullary control of muscular and hepatic glycogenolysis and of pancreatic hormonal secretion in exercising rats. Acta Physiol Scand. 1980;108:235–42.
234. Raschka C, Schuhmann R, Plath M, Parzeller M. Changes of hormone values during an ultra long distance run. In: Steinacker JM, Ward SA, editors. The physiology and pathophysiology of exercise tolerance. New York: Springer Science & Business Media; 1996.
235. American College of Sports Medicine. ACSM's guidelines for exercise testing and prescription. 10th ed. Philadelphia: Lippincott Williams & Wilkins; 2016.
236. American College of Sports Medicine. ACSM's guidelines for exercise testing and prescription. 5th ed. Philadelphia: Lippincott Williams & Wilkins; 1995.
237. Wahl P. Hormonal and metabolic responses to high intensity interval training. J Sports Med Doping Stud. 2013;3:1–2.
238. Wahl P, Zinner C, Achtzehn S, Bloch W, Mester J. Effect of high- and low-intensity exercise and metabolic acidosis on levels of GH, IGF-I, IGFBP-3 and cortisol. Growth Hormon IGF Res. 2010;20:380–5.
239. Gibala MJ, Little JP, van Essen M, Wilkin GP, Burgomaster KA, Safdar A, et al. Short-term sprint interval versus traditional endurance training: similar initial adaptations in human skeletal muscle and exercise performance. J Physiol. 2006;575:901–11.
240. Gordon SE, Kraemer WJ, Vos NH, Lynch JM, Knuttgen HG. Effect of acid-base balance on the growth hormone response to acute high-

intensity cycle exercise. J Appl Physiol (1985). 1994;76:821–9.
241. Batacan RB Jr, Duncan MJ, Dalbo VJ, Tucker PS, Fenning AS. Effects of high-intensity interval training on cardiometabolic health: a systematic review and meta-analysis of intervention studies. Br J Sports Med. 2017;51:494–503.
242. Strobel G, Friedmann B, Jost J, Bartsch P. Plasma and platelet catecholamine and catecholamine sulfate response to various exercise tests. Am J Physiol. 1994;267:E537–43.
243. Cryer PE. Physiology and pathophysiology of the human sympathoadrenal neuroendocrine system. N Engl J Med. 1980;303:436–44.
244. Kindermann W, Schnabel A, Schmitt WM, Biro G, Cassens J, Weber F. Catecholamines, growth hormone, cortisol, insulin, and sex hormones in anaerobic and aerobic exercise. Eur J Appl Physiol Occup Physiol. 1982;49:389–99.
245. Pullinen T, Mero A, MacDonald E, Pakarinen A, Komi PV. Plasma catecholamine and serum testosterone responses to four units of resistance exercise in young and adult male athletes. Eur J Appl Physiol Occup Physiol. 1998;77:413–20.
246. Justice TD, Hammer GL, Davey RJ, Paramalingam N, Guelfi KJ, Lewis L, et al. Effect of antecedent moderate-intensity exercise on the glycemia-increasing effect of a 30-sec maximal sprint: a sex comparison. Physiol Rep. 2015;3:1–10.
247. Vierck J, O'Reilly B, Hossner K, Antonio J, Byrne K, Bucci L, Dodson M. Satellite cell regulation following myotrauma caused by resistance exercise. Cell Biol Int. 2000;24:263–72.
248. Galbo H. Endocrine factors in endurance. In: Shepard RJ, Astrand PO, editors. Endurance in sport. Oxford: Blackwell Scientific Publications; 1992.
249. Philippou A, Maridaki M, Pneumaticos S, Koutsilieris M. The complexity of the IGF1 gene splicing, posttranslational modification and bioactivity. Mol Med. 2014;20:202–14.
250. Eliakim A, Nemet D, Most G, Rakover N, Pantanowitz M, Meckel Y. Effect of gender on the GH-IGF-I response to anaerobic exercise in young adults. J Strength Cond Res. 2014;28:3411–5.
251. Philippou A, Papageorgiou E, Bogdanis G, Halapas A, Sourla A, Maridaki M, et al. Expression of IGF-1 isoforms after exercise-induced muscle damage in humans: characterization of the MGF E peptide actions in vitro. In Vivo. 2009;23:567–75.
252. Gatti R, De Palo EF, Antonelli G, Spinella P. IGF-I/IGFBP system: metabolism outline and physical exercise. J Endocrinol Investig. 2012;35:699–707.
253. Lovell DI, Cuneo R, Wallace J, McLellan C. The hormonal response of older men to sub-maximum aerobic exercise: the effect of training and detraining. Steroids. 2012;77:413–8.
254. Hartgens F, Kuipers H. Effects of androgenic-anabolic steroids in athletes. Sports Med. 2004;34:513–54.
255. Tamaki T, Uchiyama S, Uchiyama Y, Akatsuka A, Roy RR, Edgerton VR. Anabolic steroids increase exercise tolerance. Am J Physiol Endocrinol Metab. 2001;280:E973–81.
256. Hackney AC, Hosick KP, Myer A, Rubin DA, Battaglini CL. Testosterone responses to intensive interval versus steady-state endurance exercise. J Endocrinol Invest. 2012;35:947–50.
257. Kraemer WJ, Gordon SE, Fleck SJ, Marchitelli LJ, Mello R, Dziados JE, et al. Endogenous anabolic hormonal and growth factor responses to heavy resistance exercise in males and females. Int J Sports Med. 1991;12:228–35.
258. Pullinen T, Mero A, Huttunen P, Pakarinen A, Komi PV. Resistance exercise-induced hormonal responses in men, women, and pubescent boys. Med Sci Sports Exerc. 2002;34:806–13.
259. Davis SN, Galassetti P, Wasserman DH, Tate D. Effects of gender on neuroendocrine and metabolic counterregulatory responses to exercise in normal man. J Clin Endocrinol Metab. 2000;85:224–30.
260. Berg U, Enqvist JK, Mattsson CM, Carlsson-Skwirut C, Sundberg CJ, Ekblom B, et al. Lack of sex differences in the IGF-IGBP response to ultra endurance exercise. Scand J Med Sci Sports. 2008;18:706–14.
261. Horton TJ, Grunwald GK, Lavely J, Donahoo WT. Glucose kinetics differ between women and men, during and after exercise. J Appl Physiol (1985). 2006;100:1883–94.
262. Buckley J. Endocrine factors in endurance. In: Maclaren D, Spurway N, editors. Exercise physiology in special populations. Advances in sport and exercise science. Livingstone: Churchill; 2008.
263. Pedersen BK, Saltin B. Exercise as medicine – evidence for prescribing exercise as therapy in 26 different chronic diseases. Scand J Med Sci Sports. 2015;25(Suppl 3):1–72.
264. Yardley JE, Hay J, Abou-Setta AM, Marks SD, McGavock J. A systematic review and meta-analysis of exercise interventions in adults with type 1 diabetes. Diabetes Res Clin Pract. 2014;106:393–400.
265. Krentz AJ, Bailey CJ. Type 2 diabetes in practice. London: Royal Society of Medicine Press; 2001.
266. Kokkinos P, Faselis C, Narayan P, Myers J, Nylen E, Sui X, Zhang J, Lavie CJ. Cardiorespiratory fitness and incidence of type 2 diabetes in united states veterans on statin therapy. Am J Med. 2017;130:1192–8.
267. Kokkinos P. Physical activity, health benefits, and mortality risk. ISRN Cardiol. 2012;2012:718789.
268. Lavie CJ, De Schutter A, Parto P, Jahangir E, Kokkinos P, Ortega FB, et al. Obesity and prevalence of cardiovascular diseases and prognosis-the obesity paradox updated. Prog Cardiovasc Dis. 2016;58:537–47.
269. Nylen ES, Gandhi SM, Kheirbek R, Kokkinos P. Enhanced fitness and renal function in type 2 diabetes. Diabet Med. 2015;32:1342–5.

270. Kokkinos P, Sheriff H, Kheirbek R. Physical inactivity and mortality risk. Cardiol Res Pract. 2010;2011:924945.
271. Goff DC Jr, Lloyd-Jones DM, Bennett G, Coady S, D'Agostino RB, Gibbons R, et al. 2013 ACC/AHA Guideline on the Assessment of Cardiovascular Risk: a report of the American College of Cardiology/American Heart Association Task Force on Practice Guidelines. Circulation. 2013;129:S49–73.
272. Nylen E, Ni D, Myersb J, Manchin C, Plunkett M, Kokkinos P. Cardiorespiratory fitness impact on all-cause mortality in prediabetic veterans. J Endocrinol Metab. 2015;5:215–9.
273. Dela F, von Linstow ME, Mikines KJ, Galbo H. Physical training may enhance beta-cell function in type 2 diabetes. Am J Physiol Endocrinol Metab. 2004;287:E1024–31.
274. Hawley JA. Exercise as a therapeutic intervention for the prevention and treatment of insulin resistance. Diabetes Metab Res Rev. 2004;20:383–93.
275. Colberg SR, Sigal RJ, Yardley JE, Riddell MC, Dunstan DW, Dempsey PC, et al. Physical activity/exercise and diabetes: a position statement of the american diabetes association. Diabetes Care. 2016;39:2065–79.
276. Kerksick CM, Fox E. Sports nutrition needs for child and adolescent athletes. Boca Raton: CRC Press; 2016.
277. Reyes LM, Davenport MH. Exercise as a therapeutic intervention to optimize fetal weight. Pharmacol Res. 2018;132:160–7.
278. Gallen IW. Exercise for people with type 1 diabetes. Med Sport Sci. 2014;60:141–53.
279. Moser O, Yardley JE, Bracken RM. Interstitial glucose and physical exercise in type 1 diabetes: integrative physiology, technology, and the gap in-between. Nutrients. 2018;10:1–15.
280. MacDonald MJ. Postexercise late-onset hypoglycemia in insulin-dependent diabetic patients. Diabetes Care. 1987;10:584–8.
281. Gallen IW, Hume C, Lumb A. Fuelling the athlete with type 1 diabetes. Diabetes Obes Metab. 2011;13:130–6.
282. Guelfi KJ, Jones TW, Fournier PA. New insights into managing the risk of hypoglycaemia associated with intermittent high-intensity exercise in individuals with type 1 diabetes mellitus: implications for existing guidelines. Sports Med. 2007;37:937–46.
283. Yardley JE, Sigal RJ. Exercise strategies for hypoglycemia prevention in individuals with type 1 diabetes. Diabetes Spectr. 2015;28:32–8.
284. MacLeod SF, Terada T, Chahal BS, Boule NG. Exercise lowers postprandial glucose but not fasting glucose in type 2 diabetes: a meta-analysis of studies using continuous glucose monitoring. Diabetes Metab Res Rev. 2013;29:593–603.
285. Colberg SR, Albright AL, Blissmer BJ, Braun B, Chasan-Taber L, Fernhall B, et al. Exercise and type 2 diabetes: American college of sports medicine and the american diabetes association: joint position statement. Exercise and type 2 diabetes. Med Sci Sports Exerc. 2010;42:2282–303.
286. De Nardi AT, Tolves T, Lenzi TL, Signori LU, Silva A. High-intensity interval training versus continuous training on physiological and metabolic variables in prediabetes and type 2 diabetes: a meta-analysis. Diabetes Res Clin Pract. 2018;137:149–59.
287. Lee S, Kim Y. Effects of exercise alone on insulin sensitivity and glucose tolerance in obese youth. Diabetes Metab J. 2013;37:225–32.
288. Church TS, Blair SN, Cocreham S, Johannsen N, Johnson W, Kramer K, Mikus CR, Myers V, Nauta M, Rodarte RQ, Sparks L, Thompson A, et al. Effects of aerobic and resistance training on hemoglobin A1c levels in patients with type 2 diabetes: a randomized controlled trial. JAMA. 2010;304:2253–62.
289. Umpierre D, Ribeiro PA, Kramer CK, Leitao CB, Zucatti AT, Azevedo MJ, et al. Physical activity advice only or structured exercise training and association with HbA1c levels in type 2 diabetes: a systematic review and meta-analysis. JAMA. 2011;305:1790–9.
290. Kwak HB. Exercise and obesity-induced insulin resistance in skeletal muscle. Integr Med Res. 2013;2:131–8.
291. Plomgaard P, Weigert C. Do diabetes and obesity affect the metabolic response to exercise? Curr Opin Clin Nutr Metab Care. 2017;20:294–9.
292. Kopin L, Lowenstein C. Dyslipidemia. Ann Intern Med. 2017;167:ITC81–96.
293. Leon AS, Sanchez OA. Response of blood lipids to exercise training alone or combined with dietary intervention. Med Sci Sports Exerc. 2001;33:S502–15; discussion S528–509
294. Breneman CB, Polinski K, Sarzynski MA, Lavie CJ, Kokkinos PF, Ahmed A, et al. The impact of cardiorespiratory fitness levels on the risk of developing atherogenic dyslipidemia. Am J Med. 2016;129:1060–6.
295. Sui X, Sarzynski MA, Lee DC, Kokkinos PF. Impact of changes in cardiorespiratory fitness on hypertension, dyslipidemia and survival: An overview of the epidemiological evidence. Prog Cardiovasc Dis. 2017;60:56–66.
296. Hayashino Y, Jackson JL, Fukumori N, Nakamura F, Fukuhara S. Effects of supervised exercise on lipid profiles and blood pressure control in people with type 2 diabetes mellitus: a meta-analysis of randomized controlled trials. Diabetes Res Clin Pract. 2012;98:349–60.
297. Mann S, Beedie C, Jimenez A. Differential effects of aerobic exercise, resistance training and combined exercise modalities on cholesterol and the lipid profile: review, synthesis and recommendations. Sports Med. 2014;44:211–21.
298. Kodama S, Tanaka S, Saito K, Shu M, Sone Y, Onitake F, Suzuki E, Shimano H, Yamamoto S, Kondo K, Ohashi Y, Yamada N, Sone H. Effect of aerobic exercise training on serum levels of high-density lipoprotein cholesterol: a meta-analysis. Arch Intern Med. 2007;167:999–1008.

299. Pronk NP. Short term effects of exercise on plasma lipids and lipoproteins in humans. Sports Med. 1993;16:431–48.
300. Kokkinos PF, Holland JC, Narayan P, Colleran JA, Dotson CO, Papademetriou V. Miles run per week and high-density lipoprotein cholesterol levels in healthy, middle-aged men. A dose-response relationship. Arch Intern Med. 1995;155:415–20.
301. Kraus WE, Houmard JA, Duscha BD, Knetzger KJ, Wharton MB, McCartney JS, et al. Effects of the amount and intensity of exercise on plasma lipoproteins. N Engl J Med. 2002;347:1483–92.
302. Mestek ML, Plaisance EP, Ratcliff LA, Taylor JK, Wee SO, Grandjean PW. Aerobic exercise and postprandial lipemia in men with the metabolic syndrome. Med Sci Sports Exerc. 2008;40:2105–11.
303. Pafili ZK, Bogdanis GC, Tsetsonis NV, Maridaki M. Postprandial lipemia 16 and 40 hours after low-volume eccentric resistance exercise. Med Sci Sports Exerc. 2009;41:375–82.
304. Sabaka P, Kruzliak P, Balaz D, Komornikova A, Celovska D, Cammarota G, et al. Effect of short term aerobic exercise on fasting and postprandial lipoprotein subfractions in healthy sedentary men. Lipids Health Dis. 2015;14:151.
305. Kokkinos PF, Narayan P, Colleran J, Fletcher RD, Lakshman R, Papademetriou V. Effects of moderate intensity exercise on serum lipids in African-American men with severe systemic hypertension. Am J Cardiol. 1998;81:732–5.
306. Durstine JL, Grandjean PW, Cox CA, Thompson PD. Lipids, lipoproteins, and exercise. J Cardiopulm Rehabil. 2002;22:385–98.
307. Quinlivan R, Buckley J, James M, Twist A, Ball S, Duno M, et al. McArdle disease: a clinical review. J Neurol Neurosurg Psychiatry. 2010;81:1182–8.
308. Mc AB. Myopathy due to a defect in muscle glycogen breakdown. Clin Sci. 1951;10:13–35.
309. Nogales-Gadea G, Santalla A, Ballester-Lopez A, Arenas J, Martin MA, Godfrey R, et al. Exercise and preexercise nutrition as treatment for McArdle disease. Med Sci Sports Exerc. 2015;48:673–9.
310. Delaney NF, Sharma R, Tadvalkar L, Clish CB, Haller RG, Mootha VK. Metabolic profiles of exercise in patients with McArdle disease or mitochondrial myopathy. Proc Natl Acad Sci USA. 2017;114:8402–7.
311. Nogales-Gadea G, Godfrey R, Santalla A, Coll-Canti J, Pintos-Morell G, Pinos T, et al. Genes and exercise intolerance: Insights from McArdle disease. Physiol Genomics. 2016;48:93–100.
312. Kaczor JJ, Robertshaw HA, Tarnopolsky MA. Higher oxidative stress in skeletal muscle of McArdle disease patients. Mol Genet Metab Rep. 2017;12:69–75.
313. Quinlivan R, Martinuzzi A, Schoser B. Pharmacological and nutritional treatment for McArdle disease (glycogen storage disease type v). Cochrane Database Syst Rev. 2014;11:CD003458.
314. Santalla A, Nogales-Gadea G, Ortenblad N, Brull A, de Luna N, Pinos T, et al. McArdle disease: a unique study model in sports medicine. Sports Med. 2014;44:1531–44.
315. Nogales-Gadea G, Consuegra-Garcia I, Rubio JC, Arenas J, Cuadros M, Camara Y, et al. A transcriptomic approach to search for novel phenotypic regulators in McArdle disease. PLoS One. 2012;7:e31718.
316. Munguia-Izquierdo D, Santalla A, Lucia A. Cardiorespiratory fitness, physical activity, and quality of life in patients with McArdle disease. Med Sci Sports Exerc. 2015;47:799–808.
317. Ollivier K, Hogrel JY, Gomez-Merino D, Romero NB, Laforet P, Eymard B, et al. Exercise tolerance and daily life in McArdle's disease. Muscle Nerve. 2005;31:637–41.
318. Andersen ST, Haller RG, Vissing J. Effect of oral sucrose shortly before exercise on work capacity in McArdle disease. Arch Neurol. 2008;65:786–9.
319. Mate-Munoz JL, Moran M, Perez M, Chamorro-Vina C, Gomez-Gallego F, Santiago C, et al. Favorable responses to acute and chronic exercisein McArdle patients. Clin J Sport Med. 2007;17:297–303.
320. Quinlivan R, Vissing J, Hilton-Jones D, Buckley J. Physical training for McArdle disease. Cochrane Database Syst Rev. 2011;12:CD007931.
321. Haller RG, Wyrick P, Taivassalo T, Vissing J. Aerobic conditioning: an effective therapy in McArdle's disease. Ann Neurol. 2006;59:922–8.
322. Garcia-Benitez S, Fleck SJ, Naclerio F, Martin MA, Lucia A. Resistance (weight lifting) training in an adolescent with McArdle disease. J Child Neurol. 2013;28:805–8.
323. Santalla A, Munguia-Izquierdo D, Brea-Alejo L, Pagola-Aldazabal I, Diez-Bermejo J, Fleck SJ, et al. Feasibility of resistance training in adult McArdle patients: clinical outcomes and muscle strength and mass benefits. Front Aging Neurosci. 2014;6:334.
324. Garber CE, Blissmer B, Deschenes MR, Franklin BA, Lamonte MJ, Lee IM, et al. American College of Sports Medicine Position Stand. Quantity and quality of exercise for developing and maintaining cardiorespiratory, musculoskeletal, and neuromotor fitness in apparently healthy adults: guidance for prescribing exercise. Med Sci Sports Exerc. 2011;43:1334–59.
325. Andersen ST, Vissing J. Carbohydrate- and protein-rich diets in McArdle disease: effects on exercise capacity. J Neurol Neurosurg Psychiatry. 2008;79:1359–63.
326. Reason SL, Westman EC, Godfrey R, Maguire E. Can a low-carbohydrate diet improve exercise tolerance in McArdle disease? J Rare Disord Diagn Ther. 2017;3:1–5.