**Name of Journal: *World Journal of Orthopedics***

**ESPS Manuscript NO: 27038**

**Manuscript Type: Review**

**Skeletal muscle mitochondrial health and spinal cord injury**

O’Brien LC *et al*. Mitochondria and spinal cord injury

**Laura C O’Brien, Ashraf S Gorgey**

**Laura C O’Brien, Ashraf S Gorgey**, Spinal Cord Injury and Disorders Service, Hunter Holmes McGuire VA Medical Center, Richmond, VA 23249, United States

**Laura C O’Brien**, Department of Physiology and Biophysics, Virginia Commonwealth University, Richmond, VA 23298, United States

**Ashraf S Gorgey**, Department of Physical Medicine and Rehabilitation, Virginia Commonwealth University, Richmond, VA 23298, United States

**Author contributions:** O’Brien LC and Gorgey AS wrote the paper

**Conflict-of-interest statement:** The authors declare no conflict of interest

**Open-Access:** This article is an open-access article which was selected by an in-house editor and fully peer-reviewed by external reviewers. It is distributed in accordance with the Creative Commons Attribution Non Commercial (CC BY-NC 4.0) license, which permits others to distribute, remix, adapt, build upon this work non-commercially, and license their derivative works on different terms, provided the original work is properly cited and the use is non-commercial. See: <http://creativecommons.org/licenses/by-nc/4.0/>

**Manuscript source:** Invited manuscript

**Correspondence to:** **Ashraf S Gorgey, MPT, PhD, FACSM, Chief** of Spinal Cord Injury Research, Hunter Holmes McGuire VA Medical Center Spinal Cord Injury and Disorders Service, 1201 Broad Rock Blvd, Richmond, VA 23249, United States. ashraf.gorgey@va.gov

**Telephone:** +1-804-6755000

**Fax:** +1-804-6755223

**Received:** May 5, 2016

**Peer-review started:** May 9, 2016

**First decision:** June 13, 2016

**Revised:** June 18, 2016

**Accepted:** August 15, 2016

**Article in press:**

**Published online:**

**Abstract**

Mitochondria are the main source of cellular energy production and are dynamic organelles that undergo biogenesis, remodeling, and degradation. Mitochondrial dysfunction is observed in a number of disease states including acute and chronic central or peripheral nervous system injury by traumatic brain injury, spinal cord injury (SCI), and neurodegenerative disease as well as in metabolic disturbances such as insulin resistance, type II diabetes and obesity. Mitochondrial dysfunction is most commonly observed in high energy requiring tissues like the brain and skeletal muscle. In persons with chronic SCI, changes to skeletal muscle may include remarkable atrophy and conversion of muscle fiber type from oxidative to fast glycolytic, combined with increased infiltration of intramuscular adipose tissue. These changes contribute to a proinflammatory environment, glucose intolerance and insulin resistance. The loss of metabolically active muscle combined with inactivity predisposes individuals with SCI to type II diabetes and obesity. The contribution of skeletal muscle mitochondrial density and electron transport chain activity to the development of the aforementioned comorbidities following SCI is unclear. A better understanding of the mechanisms involved in skeletal muscle mitochondrial dynamics is imperative to designing and testing effective treatments for this growing population. The current editorial will review ways to study mitochondrial function and the importance of improving skeletal muscle mitochondrial health in clinical populations with a special focus on chronic SCI.

**Key words:** Mitochondria; Spinal cord injuries; Body composition; Diabetes mellitus; Obesity; Metabolism

**© The Author(s) 2016.** Published by Baishideng Publishing Group Inc. All rights reserved.

**Core tip:** Mitochondria are the main source of cellular energy production and have decreased function in many disease states. After spinal cord injury (SCI) there is a dramatic deterioration of body composition including increased adipose tissue deposition, skeletal muscle atrophy and conversion from oxidative to glycolytic skeletal muscle fibers. These changes put persons with SCI at a high risk for developing cardiovascular disease and type II diabetes. How skeletal muscle mitochondrial function is impacted after human SCI has yet to be determined. The current editorial will discuss the importance of studying skeletal muscle mitochondrial function after SCI.

O’Brien LC, Gorgey AS. Skeletal muscle mitochondrial health and spinal cord injury. *World J Orthop* 2016; In press

**INTRODUCTION**

Mitochondria produce over 95% of ATP through the process of oxidative phosphorylation. Under physiological conditions, mitochondria undergo a dynamic process of biogenesis, remodeling and degradation. Dysregulation of this balance results in decreased energy production, increased reactive oxygen species (ROS) and in some cases cell death. Mitochondrial dysfunction is observed with normal aging, as well as in many disease states. It is well established that damage to the central nervous system (CNS) by traumatic brain injury, spinal cord injury (SCI) and neurodegenerative diseases (Alzheimer’s disease, Parkinson’s disease, Huntington’s disease and amyotrophic lateral sclerosis) is associated with mitochondrial dysfunction[1]. Recent studies have suggested that metabolic disorders including atherosclerosis, hypertension, cancer, insulin resistance, type II diabetes and obesity is associated with decreased mitochondrial function as well[2-4]. A better understanding of the mechanisms involved in mitochondrial dynamics and ways to improve mitochondrial health could be important for designing and testing effective treatments for these clinical populations. In this editorial we will discuss the importance of studying skeletal muscle mitochondrial health and function in persons with chronic SCI.

SCI is a devastating medical condition that results from direct or indirect damage to the spinal cord. This damage can be caused by trauma or by several pathological conditions. There are approximately 12000 new cases of SCI each year in the United States and nearly half of these individuals are between the ages of 16 and 30[5]. Because the near-normal life expectancy of persons with SCI, the estimated lifetime cost of health care and living expenses for a person with a cervical SCI is over $3 million, not including lost income[5]. After SCI, patients undergo severe body composition deterioration and skeletal muscle changes that predispose them to metabolic disorders like type II diabetes and cardiovascular disease[6-8].

**MITOCHONDRIAL DYNAMICS AND ENERGY PRODUCTION**

***Cellular energy production***

Mitochondria are double membraned organelles. The outer mitochondrial membrane (OMM) allows the passage of small molecules through voltage-dependent anion channels; however, access to the inner mitochondrial membrane (IMM) is much more tightly regulated[9]. The electron transport chain (ETC) consists of IMM-bound protein complexes I-IV and functions to maintain the electrochemical gradient across the IMM that is necessary to make ATP[10] (Figures 1 and 2). This electrochemical gradient is achieved by the pumping of protons (H+) from the mitochondrial matrix into the intermembrane space (IMS) by complexes I, III, and IV (Figure 2). Disruption of this gradient results in decreased ATP synthesis and causes electrons to leak from the ETC and react with molecular oxygen in the matrix to create the ROS superoxide (O2**-**)[11].

The main source of electrons for the ETC is the reduced form of nicotinamide adenine dinucleotide (NADH) produced by the citric acid (Kreb’s) cycle and the oxidation of fatty acids (β-oxidation; Figure 1). Another source of electrons is succinate, a byproduct of the Kreb’s cycle. Electrons enter the ETC through complex I (NADH dehydrogenase) or complex II (succinate dehydrogenase) and are then transferred to complex III (cytochrome bc1 complex) through a lipid soluble carrier molecule, coenzyme Q (Figure 2). Electrons then move between complex III and IV by way of a water soluble carrier molecule, cytochrome c. Molecular oxygen is reduced and water is produced by complex IV, cytochrome c oxidase. The movement of electrons through the ETC is coupled to ATP production by ATP synthase, or complex V. This protein complex converts ADP to ATP and is coupled to proton movement from the IMS back into the matrix. This process of synthesizing ATP in the mitochondria is called oxidative phosphorylation.

Electrons can leak from the ETC and react with molecular oxygen to create ROS like O2**-** (Figure 2). Complex I and III are the primary sites of electron leak and are sensitive to ROS injury [11]. ROS play an important role in skeletal muscle plasticity and activate many signaling cascades including increasing peroxisome proliferator-activated receptor gamma coactivator 1-alpha (PGC-1α), the master regulator of mitochondrial biogenesis[12,13]. Cells have antioxidants in the cytoplasm and mitochondrial matrix in order to neutralize ROS. However, if the balance between antioxidants and ROS is disturbed, large amounts of ROS can result in the oxidation of proteins, lipids and DNA. As discussed above, access across the IMM is tightly regulated and disruption of the electrochemical gradient across it results in decreased ATP synthesis and increased ROS production. In addition to mitochondrial ROS production, in skeletal muscle, superoxide is also produced in the sarcoplasmic reticulum, transverse tubules, sarcolemma, and the cytosol[14].

***Mitochondrial biogenesis***

Mitochondrial biogenesis includes transcription of genes by the nuclear and mitochondrial genomes. Mitochondrial DNA (mtDNA) is circular and encodes 13 proteins essential for ETC function. MtDNA is susceptible to damage by ROS because it is not protected by histones like nuclear DNA. Deletions in mtDNA are observed in several diseases and may result in a decrease in gene expression of mitochondrial encoded genes important for ETC function, resulting in increased ROS production and decreased ATP production.

Mitochondrial biogenesis is regulated, in part, by the master regulator PGC-1α and varies based on the energy needs of the cell. After translation and mitochondrial import, proteins of the ETC are assembled into protein complexes and generate ATP through oxidative phosphorylation. For a review on this topic[13].

***Mitochondrial dynamics***

Remodeling of mitochondria occurs by fission and fusion. Fusion of mitochondria results in a large mitochondrial network, while fission pinches off a part of this network. After fission, mitochondrial fragments can be tagged for degradation by a specialized type of autophagy, termed mitophagy, or they can rejoin the mitochondrial network by fusion. When there is a scarcity of nutrients the mitochondrial network is fused in order to increase mitochondrial bioenergetic efficiency, while an abundance of nutrients results in a fragmented mitochondrial network[15]. Mitochondria also make contact with other subcellular organelles, including the endoplasmic reticulum. These endoplasmic reticulum mitochondria-associated membranes play an important role in cellular functions including mitochondrial division, apoptosis, and lipid and calcium regulation[15,16].

**METHODS FOR STUDYING MITOCHONDRIA**

There are multiple techniques that can be used to measure mitochondrial dynamics and function. These include measuring mRNA or protein expression, enzyme activity, and oxygen consumption by respirometry. Skeletal muscle biopsies are most commonly used in human studies since spinal cord and brain tissue is inaccessible.

***mRNA and protein expression***

mRNA and protein expression of molecules involved in mitochondrial biogenesis, remodeling and degradation can be observed by quantitative (real-time) polymerase chain reaction (qPCR) and western blot. This is the easiest way to detect changes in cellular signaling cascades and allows for the elucidation of where in the cascade there is a potential defect caused by disease or injury. Western blot analysis of ETC proteins and other mitochondrial membrane proteins can be assayed as an estimate of respiratory chain function and mitochondrial mass, respectively. However, this type of analysis assumes that the proteins will then be imported into the mitochondria and properly assembled into the ETC complexes. There is also the assumption that a change in the expression of one protein from each complex is representative of the whole complex when in fact there are multiple proteins in each complex. For example, the mammalian complex I contains 45 different proteins[17].

***Enzyme activity***

One way to estimate mitochondrial function is to measure the activity of individual ETC complexes using a spectrophotometer. This can be done by measuring specific donor-acceptor oxidoreductase activities as previously described[18,19]. By using specific substrates and inhibitors each complex can be assayed individually. For example, complex I activity can be defined as rotenone sensitive NADH oxidation, complex II by succinate dehydrogenase activity, complex III by the reduction of cytochrome c and complex IV by the oxidation of cytochrome c. Linked activity between complexes can be measured by adding either NADH (for complex I) or succinate (for complex II) and measuring the reduction of cytochrome c by complex III[18,19]. Other key enzymes of the citric acid cycle (*i.e.,* citrate synthase) and β-oxidation pathways (*i.e.,* β-hydroxyacyl-CoA dehydrogenase (β-HAD)) can also be measured spectrophotometrically[18,20].

The benefit of spectrophotometric analysis is that it uses a small amount of tissue, samples can be previously frozen, and spectrophotometers are common lab equipment[19]. A limitation of spectrophotometric analysis is that it shows maximum capacity of individual complexes and does not necessarily represent physiological function. However, with the limited sample size obtained by human skeletal muscle biopsy and the need by many labs to freeze tissue, this may be a good option for many research groups.

Previous research has used human biopsy or autopsy specimens from tissues such as skeletal muscle, liver and skin[21]. Studies have been conducted using both tissue homogenates and isolated mitochondria from previously frozen human skeletal muscle[22,23]. Isolating mitochondria is ideal for understanding mechanism; however, the cellular context is lost. Also, more tissue is needed compared to running tissue homogenates because some mitochondria will be lost in the isolation process. A benefit of analyzing tissue homogenates is that the mitochondria remain in their cellular context, giving a more physiological reading. A limitation of analyzing tissue homogenates is the high non-mitochondrial NADH oxidase activity of some tissues[24].

***Respirometry***

Measuring oxygen consumption of isolated mitochondria or permeabilized cells by respirometry is currently the gold standard for assaying mitochondrial function. Respiration shows that the ETC is functioning because oxygen is necessary for ATP synthesis. The addition of substrates and inhibitors allows assessment of individual ETC complexes and coupling to ATP synthesis. Respiration can determine mitochondrial dysfunction not identified by spectrophotometric analysis, including impairment in mitochondrial membrane transport, problems with substrate utilization, and defects in fatty acid metabolism[25]. Protocols have recently been developed to analyze mitochondrial function from as little as 20-50 mg of muscle tissue[26]. However, this technique is labor intensive and samples cannot be previously frozen because freezing uncouples the ETC from ATP synthesis.

**CHANGES IN BODY COMPOSITION AND METABOLISM AFTER SCI**

SCI is usually a result of trauma to the spine, resulting in damage to cells that send messages to and from the brain. Damage can be to the upper motor neurons that project from the brain to the spinal cord or lower motor neurons that project from the spinal cord to the muscles. The location and severity of the injury largely determines the extent of impairment. Injuries resulting in motor and sensory impairment distal to the level of injury are classified as either complete or incomplete SCI, with incomplete injury resulting in spared sensation and/or motor function. The loss of peripheral nervous system control below the level of injury results in decreased mobility. This immobility, combined with hormonal changes and poor dietary habits result in decreased muscle mass and increased adipose deposition[6,7,27]. These changes put individuals with SCI at a high risk for developing cardiovascular disease, type II diabetes, and obesity[7,28,29]. Recent studies have shown a link between the deterioration in body composition and the impaired metabolic profile after SCI[30,31]. However, there are still many questions that remain unanswered at the cellular level.

***Body composition after SCI***

Drastic changes in body composition follow SCI[8]. Skeletal muscle atrophy combined with inactivity and poor diet contributes to the increased prevalence of obesity in this population[32]. Excess body fat, particularly around the waist, is a risk factor for a number of conditions including cardiovascular and metabolic disease[31,33]. Measurements of body mass index (BMI) do not take into account regional distribution of adipose tissue and underestimate fat mass in persons with SCI[34]. Waist circumference measurements do not distinguish between subcutaneous and ectopic (visceral adipose tissue (VAT)). It is important to distinguish between these two adipose tissue types, as an increase in VAT is a risk factor for cardiovascular and metabolic disease[35]. Waist circumference measurements underestimate the amount of VAT in individuals with SCI. Increased waist circumference is correlated with the amount of VAT in able bodied (AB) individuals but this is not the case in the SCI population[36]. SCI individuals have more VAT than AB individuals with the same waist circumference[37]. Collectively, these data suggest that there adipose tissue deposition is increased after SCI and that the distribution is altered compared to AB individuals. For a review on this topic[8].

In addition to increased adipose tissue disposition, individuals with SCI experience significant changes to their skeletal muscle. These changes include significant muscle atrophy, conversion of muscle fiber type from oxidative to fast glycolytic, and an increase in intramuscular fat (IMF). Both complete and incomplete SCI results in substantial atrophy of muscles below the level of injury. Incomplete SCI resulted in a 33% decrease in thigh muscle cross sectional area and an increase in IMF six weeks post-injury compared to AB controls[32]. The conversion of muscle fiber type to fast glycolytic results in an quickly fatigued muscle that can be damaged easily[38]. Additionally, an increase in fast glycolytic muscle fibers increases insulin insensitivity and may lead to diabetes[39].

As discussed above, increased VAT, but not subcutaneous adipose tissue, is a risk factor for cardiovascular disease, glucose intolerance, insulin resistance, and hyperlipidemia[35]. This may be due to the infiltration of immune cells into VAT and subsequent secretion of inflammatory cytokines including tumor necrosis factor α (TNFα), interleukin-1 β (IL-1β) and IL-6. Previous research suggests that inflammatory cytokines released by adipose tissue accelerate skeletal muscle atrophy[40-42]. A recent study investigating the interactions between adipose tissue and skeletal muscle revealed that VAT adipocytes from obese subjects decreased cultured myotube thickness and resulted in a gene expression profile suggestive of muscle atrophy[41]. The proposed mechanism is through the release of IL-1β and IL-6 and decreased insulin growth factor signaling, resulting in insulin resistance. Another type of adipose tissue that is similar to VAT, IMF, is increased after SCI and may be a contributing factor to the development of insulin resistance[32,43]. The mechanisms underlying the interplay between adipose tissue and skeletal muscle are just beginning to be understood.

***Metabolism after SCI***

In addition to changes in body composition, metabolism is disrupted after SCI. As many as 55% of individuals with SCI have metabolic syndrome, which is characterized by three or more of the following conditions: obesity, high blood pressure, insulin resistance, high triglycerides, and low high-density lipoprotein (HDL) cholesterol levels[44]. Impaired glucose tolerance was observed in 56% of persons with SCI, compared with only 18% of AB controls[45]. Individuals with SCI also have increased low-density lipoprotein (LDL) cholesterol[46,47]. These conditions worsen with age and put individuals at risk for developing cardiovascular disease and type II diabetes.

**MITOCHONDRIAL HEALTH STATUS AFTER SCI**

***CNS mitochondrial health after SCI***

The immediate damage to the spinal cord, including damage to axons and cells at the injury site is called primary injury. Models of SCI have shown an increase in intracellular sodium, chloride and calcium 15-60 min after injury[48]. An increase in intracellular calcium may result in apoptosis if the excess calcium taken up by mitochondria triggers mitochondrial permeability transition pore opening. Following this initial insult is a secondary injury, characterized by invasion of inflammatory cells and more cell death as cells invade not only the injury site, but also the spared nervous tissue. Neuronal death leads to loss of motor or sensory function and loss of oligodendrocytes leads to axonal demyelination[49].

Mitochondrial respiration in the spinal cord through complexes I and II is decreased and oxidative stress is increased at 12 and 24 h, but not after 6 h after SCI in a rat model[50]. In another study respiration and complex I and IV enzyme activity was decreased in the spinal cord after SCI[51]. In this study mitochondrial function was improved by treatment with an antioxidant. Complex I, complex IV, and pyruvate dehydrogenase are mitochondrial enzymes that are particularly vulnerable to damage by ROS and are decreased after SCI[52]. Decreasing ROS or increasing function of these enzymes may improve functional outcomes after SCI.

***Skeletal muscle mitochondrial health after SCI***

There is limited knowledge about the changes in mitochondrial function following SCI in humans. However, indirect evidence of mitochondrial function using blood flowto measure muscle oxygen consumption revealed that muscle oxidative capacity was decreased 50%-60% in participants with SCI 2.7-22 years after injury compared to AB controls[53]. A similar deficit was observed using histochemistry to measure succinate dehydrogenase (SDH) activity in muscle biopsy samples from paralyzed muscle 2-11 years post injury compared to AB controls[54,55]. In contrast, a study analyzing SDH and GAPDH activity, markers of complex II and glycolytic capacity, respectively, 6-24 wk after injury found increased activity despite greater fatigability of muscles[38]. The reason for these discrepant results in unclear, but it could be that early after injury muscle atrophy and fiber type changes results in a compensatory increase in oxidative and glycolytic enzymes but long periods of muscle inactivity result in reduced activity of oxidative and glycolytic pathways[38,54,55]. More research is needed to determine the effect of SCI on mitochondrial function.

Mitochondria are also dysfunctional in a number of metabolic diseases including type II diabetes and obesity. A large network of fused mitochondria is observed in healthy skeletal muscle, while muscle from obese and type II diabetics is fragmented[56]. Skeletal muscle mitochondrial function is decreased as well, with a 2-3 fold decrease in NADH oxidase (complex I) activity normalized to mitochondrial content in obese and type II diabetics compared to control[57].

Exercise interventions have been shown to increase skeletal muscle mitochondrial function and improve insulin sensitivity in obesity, diabetes, and aging[58,59]. Some options for exercise intervention after SCI include neuromuscular electrical stimulation-induced weight lifting and functional electrical stimulation (FES) cycling. Sixteen weeks of electrical stimulation-induced resistance training increased muscle mass and improved mitochondrial function by 25% in patients with SCI[60]. FES cycling has also been shown to increase mitochondrial function in patients with SCI. Eight weeks of FES cycling resulted in an increase in citrate synthase, a marker of mitochondrial mass[61]. Similarly, studies found increased citrate synthase as well as increased function of enzymes involved in glycolysis and β-oxidation[62,63]. Finally, SDH was increased after 4 wk of training, suggesting that complex II activity is increased with exercise[64]. Similarly, a recent study showed that a single session of low frequency electrical stimulation increased genes involved in muscle metabolism, including PGC-1α[65]. Collectively, these studies suggest that paralyzed skeletal muscle is malleable and can increase mitochondrial function in response to exercise. Additionally, IMF has been shown to decrease after resistance exercise training[66]. This would provide additional benefit to skeletal muscle and may improve insulin sensitivity.

**SIGNIFICANCE AND FUTURE DIRECTIONS**

Mitochondria are vital for energy production and play a role in a number of cellular processes including cell signaling, cell cycle progression, calcium regulation and cell death. These organelles are dynamic, and undergo changes in activity and number in response to cellular energy needs. A decrease in neuron and skeletal muscle mitochondrial function is observed in a number of disease and injury states including CNS trauma, neurodegenerative disease, type II diabetes and obesity[1-3]. However, we know very little about mitochondrial function in patients with chronic SCI. We are just beginning to understand the role of mitochondria in insulin resistance and how skeletal muscle mitochondrial function is disrupted in patients with SCI. Future research needs to be done using functional assays to assess activity of individual ETC complexes, as well as its coupling to ATP synthesis.

Increasing mitochondrial function by pharmacological activation of mitochondrial biogenesis is an active area of research[67]. There are a number of FDA approved medications as well as naturally occurring substances that activate mitochondrial biogenesis. For example, resveratrol, which is found in red wine, activates sirtuin 1 (SIRT1) and increases PGC-1α activity and mitochondrial function and was shown to improve insulin resistance in diabetic patients[68,69]. Small molecules that activate SIRT1 with improved bioavailability and potency have been developed and are currently being tested in humans. FDA approved pharmacological activators of mitochondrial biogenesis include the β2-adrenergic receptor agonist formoterol[70], the anti-diabetic drug metformin[71], the phosphodiesterase inhibitor sildenafil[72], the PPARγ agonist rosiglitazone[73], the mitochondrial permeability transition pore inhibitor cyclosporine A[74], and the angiotensin-converting enzyme inhibitor captropril[75], among others. Although these compounds are thought to exert their effects at least in part by increasing mitochondrial biogenesis, there are currently no specific activators of mitochondrial biogenesis. Future studies need to investigate the safety and efficacy of systemically increasing mitochondrial biogenesis, as well as optimizing dosing in order to maximize the therapeutic benefit.

In order to study mitochondrial function after disease or injury or to assess the efficacy of mitochondrial targeted therapies, skeletal muscle biopsies could be used because of the inaccessibility of the brain and spinal cord in humans. However, recent studies have suggested that the bioenergetic profile of blood cells is associated with physical function and inflammation as well[76,77]. Indeed, mitochondrial dysfunction is seen in blood from patients with a number of diseases including neurodegenerative diseases and type II diabetes[78,79]. Peripheral blood mononuclear cells from patients with type II diabetes and chronic kidney disease have increased inflammatory cytokines, decreased mitochondrial function and increased ROS production[80]. These studies suggest that blood cell bioenergetics may predict systemic mitochondrial function and may act as biomarkers for metabolic stress and surrogate markers for the severity of disease progression and the efficacy of therapeutics[80,81]. This represents an intriguing possibility, as obtaining blood samples are much less invasive than biopsies and could be taken more frequently in order to better characterize the time course of therapeutic intervention.

There are a number of different techniques for analyzing cellular signaling pathways and mitochondrial function. Researchers should carefully weigh the convenience of non-invasive techniques with the mechanistic detail provided by analyzing biopsy tissue. If choosing to analyze biopsy tissue, care should be taken to obtain the proper amount of tissue required for the assay and to prepare it properly in order to preserve mitochondrial function. For samples that need to be frozen, spectrophotometric analysis may be the best option for analyzing mitochondrial function, while respiration will be ideal for fresh tissue samples. Another research consideration is whether or not to isolate mitochondria and this may depend on the sample size.

As discussed above, both resistance training and FES cycling has been shown to increase mitochondrial function in persons with SCI. In addition, electrical stimulation-induced resistance training reduced VAT and IMF and increased insulin sensitivity while increasing muscle mass[66]. FES cycling has been shown to improve insulin sensitivity as well, but the effect on muscle size and body composition were minimal to modest[82]. It is unknown if conditioning the muscles with resistance training prior to FES cycling would result in greater mitochondrial and metabolic outcomes.

**CONCLUSION**

There is limited knowledge regarding skeletal muscle mitochondrial health following SCI. Challenges may stem from difficulties in capturing muscle biopsies and running biochemical analysis to determine mitochondrial mass or activity by spectroscopy or respiration. Non-invasive procedures like near-infrared spectroscopy may be available to reflect mitochondrial activity; however, mechanistic dysfunctions relevant to the involvement of different complexes may be limited.

A better understanding of how mitochondrial function is impacted in patients with chronic SCI is critical for developing interventions to increase mitochondrial function and improve metabolic outcomes. Skeletal muscle or blood cell bioenergetics may predict overall mitochondrial health and therefore be a surrogate marker of disease progression and treatment efficacy. Increasing mitochondrial function immediately following SCI may decrease cell death and improve functional outcomes. Improvement in mitochondrial function by exercise or pharmacological interventions in chronic SCI may decrease comorbidities. This will result in better health for patients and a lower financial burden for their health care. A better understanding of mitochondrial biology may also translate to a number of other diseases in which mitochondrial are dysfunctional, particularly insulin resistance, type II diabetes, and obesity.

**ACKNOWLEDGMENTS**

We would like to thank the Hunter Holmes McGuire VA Medical Center and Virginia Commonwealth University for providing the environment to conduct clinical research.

**REFERENCES**

1 **Itoh K**, Nakamura K, Iijima M, Sesaki H. Mitochondrial dynamics in neurodegeneration. *Trends Cell Biol* 2013; **23**: 64-71 [PMID: 23159640 DOI: 10.1016/j.tcb.2012.10.006]

2 **Chan DC**. Mitochondria: dynamic organelles in disease, aging, and development. *Cell* 2006; **125**: 1241-1252 [PMID: 16814712 DOI: 10.1016/j.cell.2006.06.010]

3 **Zhao J**, Zhang J, Yu M, Xie Y, Huang Y, Wolff DW, Abel PW, Tu Y. Mitochondrial dynamics regulates migration and invasion of breast cancer cells. *Oncogene* 2013; **32**: 4814-4824 [PMID: 23128392 DOI: 10.1038/onc.2012.494]

4 **Phielix E**, Mensink M. Type 2 diabetes mellitus and skeletal muscle metabolic function. *Physiol Behav* 2008; **94**: 252-258 [PMID: 18342897 DOI: 10.1016/j.physbeh.2008.01.020]

5 **National Spinal Cord Injury Statistical Center**. Spinal cord injury facts and figures at a glance. *J Spinal Cord Med* 2013; **36**: 1-2 [PMID: 23433327 DOI: 10.1179/1079026813Z.000000000136]

6 **Kocina P**. Body composition of spinal cord injured adults. *Sports Med* 1997; **23**: 48-60 [PMID: 9017859]

7 **Gater DR**. Obesity after spinal cord injury. *Phys Med Rehabil Clin N Am* 2007; **18**: 333-51, vii [PMID: 17543776 DOI: 10.1016/j.pmr.2007.03.004]

8 **Gorgey AS**, Dolbow DR, Dolbow JD, Khalil RK, Castillo C, Gater DR. Effects of spinal cord injury on body composition and metabolic profile - part I. *J Spinal Cord Med* 2014; **37**: 693-702 [PMID: 25001559 DOI: 10.1179/2045772314Y.0000000245]

9 **Lemasters JJ**, Holmuhamedov E. Voltage-dependent anion channel (VDAC) as mitochondrial governator--thinking outside the box. *Biochim Biophys Acta* 2006; **1762**: 181-190 [PMID: 16307870 DOI: 10.1016/j.bbadis.2005.10.006]

10 **Saraste M**. Oxidative phosphorylation at the fin de siècle. *Science* 1999; **283**: 1488-1493 [PMID: 10066163]

11 **Turrens JF**. Mitochondrial formation of reactive oxygen species. *J Physiol* 2003; **552**: 335-344 [PMID: 14561818 DOI: 10.1113/jphysiol.2003.049478]

12 **Hoppeler H**, Baum O, Lurman G, Mueller M. Molecular mechanisms of muscle plasticity with exercise. *Compr Physiol* 2011; **1**: 1383-1412 [PMID: 23733647 DOI: 10.1002/cphy.c100042]

13 **Scarpulla RC**, Vega RB, Kelly DP. Transcriptional integration of mitochondrial biogenesis. *Trends Endocrinol Metab* 2012; **23**: 459-466 [PMID: 22817841 DOI: 10.1016/j.tem.2012.06.006]

14 **Powers SK**, Ji LL, Kavazis AN, Jackson MJ. Reactive oxygen species: impact on skeletal muscle. *Compr Physiol* 2011; **1**: 941-969 [PMID: 23737208 DOI: 10.1002/cphy.c100054]

15 **Schrepfer E**, Scorrano L. Mitofusins, from Mitochondria to Metabolism. *Mol Cell* 2016; **61**: 683-694 [PMID: 26942673 DOI: 10.1016/j.molcel.2016.02.022]

16 **Vance JE**. MAM (mitochondria-associated membranes) in mammalian cells: lipids and beyond. *Biochim Biophys Acta* 2014; **1841**: 595-609 [PMID: 24316057 DOI: 10.1016/j.bbalip.2013.11.014]

17 **Carroll J**, Fearnley IM, Skehel JM, Shannon RJ, Hirst J, Walker JE. Bovine complex I is a complex of 45 different subunits. *J Biol Chem* 2006; **281**: 32724-32727 [PMID: 16950771 DOI: 10.1074/jbc.M607135200]

18 **Brass EP**, Hiatt WR, Gardner AW, Hoppel CL. Decreased NADH dehydrogenase and ubiquinol-cytochrome c oxidoreductase in peripheral arterial disease. *Am J Physiol Heart Circ Physiol* 2001; **280**: H603-H609 [PMID: 11158957]

19 **Spinazzi M**, Casarin A, Pertegato V, Salviati L, Angelini C. Assessment of mitochondrial respiratory chain enzymatic activities on tissues and cultured cells. *Nat Protoc* 2012; **7**: 1235-1246 [PMID: 22653162 DOI: 10.1038/nprot.2012.058]

20 **Morash AJ**, Kotwica AO, Murray AJ. Tissue-specific changes in fatty acid oxidation in hypoxic heart and skeletal muscle. *Am J Physiol Regul Integr Comp Physiol* 2013; **305**: R534-R541 [PMID: 23785078 DOI: 10.1152/ajpregu.00510.2012]

21 **Hoppel CL**, Kerr DS, Dahms B, Roessmann U. Deficiency of the reduced nicotinamide adenine dinucleotide dehydrogenase component of complex I of mitochondrial electron transport. Fatal infantile lactic acidosis and hypermetabolism with skeletal-cardiac myopathy and encephalopathy. *J Clin Invest* 1987; **80**: 71-77 [PMID: 3110216 DOI: 10.1172/JCI113066]

22 **Kelly NA**, Ford MP, Standaert DG, Watts RL, Bickel CS, Moellering DR, Tuggle SC, Williams JY, Lieb L, Windham ST, Bamman MM. Novel, high-intensity exercise prescription improves muscle mass, mitochondrial function, and physical capacity in individuals with Parkinson's disease. *J Appl Physiol (1985)* 2014; **116**: 582-592 [PMID: 24408997 DOI: 10.1152/japplphysiol.01277.2013]

23 **Menshikova EV**, Ritov VB, Fairfull L, Ferrell RE, Kelley DE, Goodpaster BH. Effects of exercise on mitochondrial content and function in aging human skeletal muscle. *J Gerontol A Biol Sci Med Sci* 2006; **61**: 534-540 [PMID: 16799133]

24 **Trounce IA**, Kim YL, Jun AS, Wallace DC. Assessment of mitochondrial oxidative phosphorylation in patient muscle biopsies, lymphoblasts, and transmitochondrial cell lines. *Methods Enzymol* 1996; **264**: 484-509 [PMID: 8965721]

25 **Puchowicz MA**, Varnes ME, Cohen BH, Friedman NR, Kerr DS, Hoppel CL. Oxidative phosphorylation analysis: assessing the integrated functional activity of human skeletal muscle mitochondria--case studies. *Mitochondrion* 2004; **4**: 377-385 [PMID: 16120399 DOI: 10.1016/j.mito.2004.07.004]

26 **Bharadwaj MS**, Tyrrell DJ, Lyles MF, Demons JL, Rogers GW, Molina AJ. Preparation and respirometric assessment of mitochondria isolated from skeletal muscle tissue obtained by percutaneous needle biopsy. *J Vis Exp* 2015; Epub ahead of print [PMID: 25741892 DOI: 10.3791/52350]

27 **Bauman WA**, Spungen AM. Metabolic changes in persons after spinal cord injury. *Phys Med Rehabil Clin N Am* 2000; **11**: 109-140 [PMID: 10680161]

28 **Duckworth WC**, Solomon SS, Jallepalli P, Heckemeyer C, Finnern J, Powers A. Glucose intolerance due to insulin resistance in patients with spinal cord injuries. *Diabetes* 1980; **29**: 906-910 [PMID: 7429029]

29 **Lavela SL**, Weaver FM, Goldstein B, Chen K, Miskevics S, Rajan S, Gater DR. Diabetes mellitus in individuals with spinal cord injury or disorder. *J Spinal Cord Med* 2006; **29**: 387-395 [PMID: 17044389]

30 **Gorgey AS**, Gater DR. Regional and relative adiposity patterns in relation to carbohydrate and lipid metabolism in men with spinal cord injury. *Appl Physiol Nutr Metab* 2011; **36**: 107-114 [PMID: 21326384 DOI: 10.1139/H10-091]

31 **Gorgey AS**, Mather KJ, Gater DR. Central adiposity associations to carbohydrate and lipid metabolism in individuals with complete motor spinal cord injury. *Metabolism* 2011; **60**: 843-851 [PMID: 20870252 DOI: 10.1016/j.metabol.2010.08.002]

32 **Gorgey AS**, Dudley GA. Skeletal muscle atrophy and increased intramuscular fat after incomplete spinal cord injury. *Spinal Cord* 2007; **45**: 304-309 [PMID: 16940987 DOI: 10.1038/sj.sc.3101968]

33 **Nakamura T**, Tokunaga K, Shimomura I, Nishida M, Yoshida S, Kotani K, Islam AH, Keno Y, Kobatake T, Nagai Y. Contribution of visceral fat accumulation to the development of coronary artery disease in non-obese men. *Atherosclerosis* 1994; **107**: 239-246 [PMID: 7980698]

34 **Spungen AM**, Adkins RH, Stewart CA, Wang J, Pierson RN, Waters RL, Bauman WA. Factors influencing body composition in persons with spinal cord injury: a cross-sectional study. *J Appl Physiol (1985)* 2003; **95**: 2398-2407 [PMID: 12909613 DOI: 10.1152/japplphysiol.00729.2002]

35 **Jensen MD**. Role of body fat distribution and the metabolic complications of obesity. *J Clin Endocrinol Metab* 2008; **93**: S57-S63 [PMID: 18987271 DOI: 10.1210/jc.2008-1585]

36 **Gorgey AS**, Mather KJ, Poarch HJ, Gater DR. Influence of motor complete spinal cord injury on visceral and subcutaneous adipose tissue measured by multi-axial magnetic resonance imaging. *J Spinal Cord Med* 2011; **34**: 99-109 [PMID: 21528633 DOI: 10.1179/107902610X12911165975106]

37 **Edwards LA**, Bugaresti JM, Buchholz AC. Visceral adipose tissue and the ratio of visceral to subcutaneous adipose tissue are greater in adults with than in those without spinal cord injury, despite matching waist circumferences. *Am J Clin Nutr* 2008; **87**: 600-607 [PMID: 18326597]

38 **Castro MJ**, Apple DF, Staron RS, Campos GE, Dudley GA. Influence of complete spinal cord injury on skeletal muscle within 6 mo of injury. *J Appl Physiol (1985)* 1999; **86**: 350-358 [PMID: 9887150]

39 **Simoneau JA**, Kelley DE. Altered glycolytic and oxidative capacities of skeletal muscle contribute to insulin resistance in NIDDM. *J Appl Physiol (1985)* 1997; **83**: 166-171 [PMID: 9216960]

40 **Hoppeler H**. Molecular networks in skeletal muscle plasticity. *J Exp Biol* 2016; **219**: 205-213 [PMID: 26792332 DOI: 10.1242/jeb.128207]

41 **Pellegrinelli V**, Rouault C, Rodriguez-Cuenca S, Albert V, Edom-Vovard F, Vidal-Puig A, Clément K, Butler-Browne GS, Lacasa D. Human Adipocytes Induce Inflammation and Atrophy in Muscle Cells During Obesity. *Diabetes* 2015; **64**: 3121-3134 [PMID: 25695947 DOI: 10.2337/db14-0796]

42 **Kelley DE**, Goodpaster BH. Stewing in Not-So-Good Juices: Interactions of Skeletal Muscle With Adipose Secretions. *Diabetes* 2015; **64**: 3055-3057 [PMID: 26294424 DOI: 10.2337/db15-0403]

43 **Elder CP**, Apple DF, Bickel CS, Meyer RA, Dudley GA. Intramuscular fat and glucose tolerance after spinal cord injury--a cross-sectional study. *Spinal Cord* 2004; **42**: 711-716 [PMID: 15303112 DOI: 10.1038/sj.sc.3101652]

44 **Nelson MD**, Widman LM, Abresch RT, Stanhope K, Havel PJ, Styne DM, McDonald CM. Metabolic syndrome in adolescents with spinal cord dysfunction. *J Spinal Cord Med* 2007; **30 Suppl 1**: S127-S139 [PMID: 17874698]

45 **Bauman WA**, Spungen AM. Disorders of carbohydrate and lipid metabolism in veterans with paraplegia or quadriplegia: a model of premature aging. *Metabolism* 1994; **43**: 749-756 [PMID: 8201966]

46 **Nash MS**, Mendez AJ. A guideline-driven assessment of need for cardiovascular disease risk intervention in persons with chronic paraplegia. *Arch Phys Med Rehabil* 2007; **88**: 751-757 [PMID: 17532897 DOI: 10.1016/j.apmr.2007.02.031]

47 **Bauman WA**, Spungen AM, Zhong YG, Rothstein JL, Petry C, Gordon SK. Depressed serum high density lipoprotein cholesterol levels in veterans with spinal cord injury. *Paraplegia* 1992; **30**: 697-703 [PMID: 1448297 DOI: 10.1038/sc.1992.136]

48 **LoPachin RM**, Gaughan CL, Lehning EJ, Kaneko Y, Kelly TM, Blight A. Experimental spinal cord injury: spatiotemporal characterization of elemental concentrations and water contents in axons and neuroglia. *J Neurophysiol* 1999; **82**: 2143-2153 [PMID: 10561394]

49 **Rabchevsky AG**, Patel SP, Springer JE. Pharmacological interventions for spinal cord injury: where do we stand? How might we step forward? *Pharmacol Ther* 2011; **132**: 15-29 [PMID: 21605594 DOI: 10.1016/j.pharmthera.2011.05.001]

50 **Sullivan PG**, Krishnamurthy S, Patel SP, Pandya JD, Rabchevsky AG. Temporal characterization of mitochondrial bioenergetics after spinal cord injury. *J Neurotrauma* 2007; **24**: 991-999 [PMID: 17600515 DOI: 10.1089/neu.2006.0242]

51 **Patel SP**, Sullivan PG, Pandya JD, Goldstein GA, VanRooyen JL, Yonutas HM, Eldahan KC, Morehouse J, Magnuson DS, Rabchevsky AG. N-acetylcysteine amide preserves mitochondrial bioenergetics and improves functional recovery following spinal trauma. *Exp Neurol* 2014; **257**: 95-105 [PMID: 24805071 DOI: 10.1016/j.expneurol.2014.04.026]

52 **McEwen ML**, Sullivan PG, Rabchevsky AG, Springer JE. Targeting mitochondrial function for the treatment of acute spinal cord injury. *Neurotherapeutics* 2011; **8**: 168-179 [PMID: 21360236 DOI: 10.1007/s13311-011-0031-7]

53 **Erickson ML**, Ryan TE, Young HJ, McCully KK. Near-infrared assessments of skeletal muscle oxidative capacity in persons with spinal cord injury. *Eur J Appl Physiol* 2013; **113**: 2275-2283 [PMID: 23703066 DOI: 10.1007/s00421-013-2657-0]

54 **Martin TP**, Stein RB, Hoeppner PH, Reid DC. Influence of electrical stimulation on the morphological and metabolic properties of paralyzed muscle. *J Appl Physiol* (1985) 1992; **72**: 1401-1406 [PMID: 1534322]

55 **Grimby G**, Broberg C, Krotkiewska I, Krotkiewski M. Muscle fiber composition in patients with traumatic cord lesion. *Scand J Rehabil Med* 1976; **8**: 37-42 [PMID: 132700]

56 **Kelley DE**, He J, Menshikova EV, Ritov VB. Dysfunction of mitochondria in human skeletal muscle in type 2 diabetes. *Diabetes* 2002; **51**: 2944-2950 [PMID: 12351431]

57 **Ritov VB**, Menshikova EV, Azuma K, Wood R, Toledo FG, Goodpaster BH, Ruderman NB, Kelley DE. Deficiency of electron transport chain in human skeletal muscle mitochondria in type 2 diabetes mellitus and obesity. *Am J Physiol Endocrinol Metab* 2010; **298**: E49-E58 [PMID: 19887598 DOI: 10.1152/ajpendo.00317.2009]

58 **Toledo FG**, Goodpaster BH. The role of weight loss and exercise in correcting skeletal muscle mitochondrial abnormalities in obesity, diabetes and aging. *Mol Cell Endocrinol* 2013; **379**: 30-34 [PMID: 23792186 DOI: 10.1016/j.mce.2013.06.018]

59 **Lanza IR**, Nair KS. Muscle mitochondrial changes with aging and exercise. *Am J Clin Nutr* 2009; **89**: 467S-471S [PMID: 19056588 DOI: 10.3945/ajcn.2008.26717D]

60 **Ryan TE**, Brizendine JT, Backus D, McCully KK. Electrically induced resistance training in individuals with motor complete spinal cord injury. *Arch Phys Med Rehabil* 2013; **94**: 2166-2173 [PMID: 23816921 DOI: 10.1016/j.apmr.2013.06.016]

61 **Chilibeck PD**, Bell G, Jeon J, Weiss CB, Murdoch G, MacLean I, Ryan E, Burnham R. Functional electrical stimulation exercise increases GLUT-1 and GLUT-4 in paralyzed skeletal muscle. *Metabolism* 1999; **48**: 1409-1413 [PMID: 10582549]

62 **Kjaer M**, Mohr T, Biering-Sørensen F, Bangsbo J. Muscle enzyme adaptation to training and tapering-off in spinal-cord-injured humans. *Eur J Appl Physiol* 2001; **84**: 482-486 [PMID: 11417439]

63 **Crameri RM**, Weston A, Climstein M, Davis GM, Sutton JR. Effects of electrical stimulation-induced leg training on skeletal muscle adaptability in spinal cord injury. *Scand J Med Sci Sports* 2002; **12**: 316-322 [PMID: 12383078]

64 **Rochester L**, Barron MJ, Chandler CS, Sutton RA, Miller S, Johnson MA. Influence of electrical stimulation of the tibialis anterior muscle in paraplegic subjects. 2. Morphological and histochemical properties. *Paraplegia* 1995; **33**: 514-522 [PMID: 8524604 DOI: 10.1038/sc.1995.112]

65 **Petrie M**, Suneja M, Shields RK. Low-frequency stimulation regulates metabolic gene expression in paralyzed muscle. *J Appl Physiol* (1985) 2015; **118**: 723-731 [PMID: 25635001 DOI: 10.1152/japplphysiol.00628.2014]

66 **Gorgey AS**, Mather KJ, Cupp HR, Gater DR. Effects of resistance training on adiposity and metabolism after spinal cord injury. *Med Sci Sports Exerc* 2012; **44**: 165-174 [PMID: 21659900 DOI: 10.1249/MSS.0b013e31822672aa]

67 **Whitaker RM**, Corum D, Beeson CC, Schnellmann RG. Mitochondrial Biogenesis as a Pharmacological Target: A New Approach to Acute and Chronic Diseases. *Annu Rev Pharmacol Toxicol* 2016; **56**: 229-249 [PMID: 26566156 DOI: 10.1146/annurev-pharmtox-010715-103155]

68 **Lagouge M**, Argmann C, Gerhart-Hines Z, Meziane H, Lerin C, Daussin F, Messadeq N, Milne J, Lambert P, Elliott P, Geny B, Laakso M, Puigserver P, Auwerx J. Resveratrol improves mitochondrial function and protects against metabolic disease by activating SIRT1 and PGC-1alpha. *Cell* 2006; **127**: 1109-1122 [PMID: 17112576 DOI: 10.1016/j.cell.2006.11.013]

69 **Brasnyó P**, Molnár GA, Mohás M, Markó L, Laczy B, Cseh J, Mikolás E, Szijártó IA, Mérei A, Halmai R, Mészáros LG, Sümegi B, Wittmann I. Resveratrol improves insulin sensitivity, reduces oxidative stress and activates the Akt pathway in type 2 diabetic patients. *Br J Nutr* 2011; **106**: 383-389 [PMID: 21385509 DOI: 10.1017/S0007114511000316]

70 **Wills LP**, Trager RE, Beeson GC, Lindsey CC, Peterson YK, Beeson CC, Schnellmann RG. The β2-adrenoceptor agonist formoterol stimulates mitochondrial biogenesis. *J Pharmacol Exp Ther* 2012; **342**: 106-118 [PMID: 22490378 DOI: 10.1124/jpet.112.191528]

71 **Kukidome D**, Nishikawa T, Sonoda K, Imoto K, Fujisawa K, Yano M, Motoshima H, Taguchi T, Matsumura T, Araki E. Activation of AMP-activated protein kinase reduces hyperglycemia-induced mitochondrial reactive oxygen species production and promotes mitochondrial biogenesis in human umbilical vein endothelial cells. *Diabetes* 2006; **55**: 120-127 [PMID: 16380484]

72 **Whitaker RM**, Wills LP, Stallons LJ, Schnellmann RG. cGMP-selective phosphodiesterase inhibitors stimulate mitochondrial biogenesis and promote recovery from acute kidney injury. *J Pharmacol Exp Ther* 2013; **347**: 626-634 [PMID: 24042162 DOI: 10.1124/jpet.113.208017]

73 **Pardo R**, Enguix N, Lasheras J, Feliu JE, Kralli A, Villena JA. Rosiglitazone-induced mitochondrial biogenesis in white adipose tissue is independent of peroxisome proliferator-activated receptor γ coactivator-1α. *PLoS One* 2011; **6**: e26989 [PMID: 22087241 DOI: 10.1371/journal.pone.0026989]

74 **Osman MM**, Lulic D, Glover L, Stahl CE, Lau T, van Loveren H, Borlongan CV. Cyclosporine-A as a neuroprotective agent against stroke: its translation from laboratory research to clinical application. *Neuropeptides* 2011; **45**: 359-368 [PMID: 21592568 DOI: 10.1016/j.npep.2011.04.002]

75 **Yanagishita T**, Tomita M, Itoh S, Mukae S, Arata H, Ishioka H, Geshi E, Konno N, Katagiri T. Protective effect of captopril on ischemic myocardium. *Jpn Circ J* 1997; **61**: 161-169 [PMID: 9070972]

76 **Tyrrell DJ**, Bharadwaj MS, Van Horn CG, Kritchevsky SB, Nicklas BJ, Molina AJ. Respirometric Profiling of Muscle Mitochondria and Blood Cells Are Associated With Differences in Gait Speed Among Community-Dwelling Older Adults. *J Gerontol A Biol Sci Med Sci* 2015; **70**: 1394-1399 [PMID: 25030980 DOI: 10.1093/gerona/glu096]

77 **Tyrrell DJ**, Bharadwaj MS, Van Horn CG, Marsh AP, Nicklas BJ, Molina AJ. Blood-cell bioenergetics are associated with physical function and inflammation in overweight/obese older adults. *Exp Gerontol* 2015; **70**: 84-91 [PMID: 26226578 DOI: 10.1016/j.exger.2015.07.015]

78 **Zharikov S**, Shiva S. Platelet mitochondrial function: from regulation of thrombosis to biomarker of disease. *Biochem Soc Trans* 2013; **41**: 118-123 [PMID: 23356269 DOI: 10.1042/BST20120327]

79 **Ladd AC**, Keeney PM, Govind MM, Bennett JP. Mitochondrial oxidative phosphorylation transcriptome alterations in human amyotrophic lateral sclerosis spinal cord and blood. *Neuromolecular Med* 2014; **16**: 714-726 [PMID: 25081190 DOI: 10.1007/s12017-014-8321-y]

80 **Ravi S**, Mitchell T, Kramer PA, Chacko B, Darley-Usmar VM. Mitochondria in monocytes and macrophages-implications for translational and basic research. *Int J Biochem Cell Biol* 2014; **53**: 202-207 [PMID: 24863362 DOI: 10.1016/j.biocel.2014.05.019]

81 **Chacko BK**, Kramer PA, Ravi S, Benavides GA, Mitchell T, Dranka BP, Ferrick D, Singal AK, Ballinger SW, Bailey SM, Hardy RW, Zhang J, Zhi D, Darley-Usmar VM. The bioenergetic health index: A new concept in mitochondrial translational research. *Clin Sci* (Lond) 2014; **127**: 367-373 [PMID: 24895057 DOI: 10.1042/CS20140101]

82 **Gorgey AS**, Dolbow DR, Dolbow JD, Khalil RK, Gater DR. The effects of electrical stimulation on body composition and metabolic profile after spinal cord injury--Part II. *J Spinal Cord Med* 2015; **38**: 23-37 [PMID: 25001669 DOI: 10.1179/2045772314Y.0000000244]

**P-Reviewer:** Berra LV, Rabchevsky AG **S-Editor:** Qiu S **L-Editor: E-Editor:**



**Figure 1 Cellular energy production.** In skeletal muscle, glucose enters the cell through glucose transporter type 1 or 4 (GLUT1 or GLUT4, respectively). Glucose is converted to pyruvate in the glycolysis pathway. Pyruvate is transported across the outer and inner mitochondrial membranes (OMM and IMM, respectively) and into the mitochondrial matrix where it is converted into acetyl-coA. Fatty acids undergo β-oxidation in the mitochondria, creating acetyl-coA and NADH. Acetyl-coA is utilized by the Kreb’s cycle, creating NADH and succinate which enter the electron transport chain (ETC) at complex I and II, respectively. The movement of electrons through the ETC is coupled to the production of ATP in a process called oxidative phosphorylation (OXPHOS). IMS: Innermembrane space.



**Figure 2 The electron transport chain is located on the inner mitochondrial membrane.** Electrons (e-) enter through complex I or II and are transferred to complex III by coenzyme Q (Q). Electrons then move between complex III and IV by cytochrome c (c). Electrons can leak from the chain and react with oxygen to create superoxide (O2-). The movement of electrons is coupled to the pumping of protons (H+) from the mitochondrial matrix into the innermembrane space (IMS). This gradient is then used by ATP synthase, or complex V to generate ATP.