

A Closer Look at Muscle Breakdown: An Overview of the Causes, Complications, and
Diagnostics of Rhabdomyolysis

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Abstract

Rhabdomyolysis is the rapid breakdown of myocytes in skeletal muscle. This condition can be triggered by a variety of things including crush injury, drugs, prolonged exercise, and venom. Although the causes can vary, the general pathophysiology of rhabdomyolysis is similar. Increased intracellular calcium ion levels instigate the breakdown of cellular and certain organelle membranes of muscle cells. Reactive oxygen species form, ATP is depleted, and the sarcoplasmic reticulum, which regulates muscle contraction, is also damaged. Necrosis ensues, leading to toxic intracellular components spreading throughout the body. Rhabdomyolysis can then lead to several complications including acute renal injury and heart arrhythmias, and is characterized by dark-colored urine, pain, and weakness. Diagnostic tests for certain chemicals, like myoglobin, help diagnose rhabdomyolysis.

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Overview of Rhabdomyolysis

Rhabdomyolysis is a serious condition where skeletal muscle fibers are damaged and rapidly broken down (1). This condition becomes dangerous when the previously harmless intracellular chemicals of the muscle cells are released into bodily circulation. The rapid release of chemicals such as myoglobin, creatine kinase, electrolytes, and lactate dehydrogenase, and other intracellular proteins first enter interstitial fluids at high concentrations (2). Colloid osmotic pressure takes effect as these chemicals enter nearby capillaries and then further spread throughout the body's blood vessels. The circulatory system then spreads the potentially harmful proteins/chemicals to other areas of the body, which can lead to adverse effects. Damage and death of skeletal muscle can be life threatening, as skeletal muscle constitutes 40% of the human body mass (3). Rhabdomyolysis heavily varies in its severity and can be caused by many different factors. Because of this, it is difficult to analyze the statistics of rhabdomyolysis. One evident characteristic is that rhabdomyolysis is closely correlated to critically ill trauma patients. More specifically, patients with blunt trauma or crush injuries are likely to have characteristics of rhabdomyolysis. It has been found, however, that there are several additional causes of rhabdomyolysis, such as drug use, toxins, genetic conditions, and bacterial infections, among others (4). Generally, rhabdomyolysis manifests in a trio of symptoms: muscle weakness, muscle pain, and dark red/brown colored urine (5). While these are the trademark signs of this condition, there are some additional symptoms of rhabdomyolysis such as confusion, irregular heartbeat, and sometimes seizures (2).

Normal Muscle Physiology

The muscular system is extremely important for normal, day to day bodily function. This system is responsible for bodily movement, both voluntary and involuntary. There are three main types of muscles: skeletal, smooth, and cardiac. Each type plays a pertinent role in bodily function. Cardiac muscle allows for proper movement of the heart, which powers blood flow in the circulatory system. Smooth muscle is mostly involuntary and is found mostly along the walls of hollow organs and glands. This includes digestive organs (stomach, intestines, esophagus), urinary organs (bladder), and even walls of blood vessels. These muscles help control bodily functions that are not under conscience control, like food digestion and micturition. Skeletal muscle is closely associated with the skeletal system. It is responsible for body movement, posture, and support. Tendons connect skeletal muscle groups to bones, which allows for controlled and coordinated voluntary movements. The musculoskeletal system works with the nervous system to allow people to walk, play sports, write a paper, and even sit still. Healthy bodies have several groups of functioning skeletal muscles of various sizes, shapes, and specific functions, but generally skeletal muscle consists of similar cellular structure.

Normally functioning muscles are made from muscle fibers. Muscle fibers are composed of fascicles, which are groupings of cells (6). Fascicles and muscle fibers are bound together by fasciae to ensure proper function. The individual muscle cells, also called myocytes, are generated during myogenesis from myoblasts (6). Typical skeletal muscle cells are characterized by multiple nuclei, abundant mitochondria, and high amounts of actin and myosin filaments within sarcomeres. The sarcomeres give the skeletal muscle tissue a striated appearance when observed microscopically (7). Muscle fibers have various ion channels on the plasma membrane, which facilitate the transport of ions like sodium, potassium, and calcium across

muscle membranes (2). Muscle depolarization and repolarization take place through this ion transport in this way to allow for sarcomere stimulation. This allows proper muscle contraction and movement. These various processes of muscle movement are reliant on energy (ATP). In normal physiology, the body supplies the muscles with enough energy to create proper contraction.

Muscle Contraction and Relaxation, and Calcium

One of the most important chemicals of myocytes is calcium because it is involved with normal physiological contraction of muscles. In skeletal muscle, the body signals a muscle group to move through nerve impulses. The nervous signals translate to muscle tissue through neuromuscular junctions. This is where neurotransmitter acetylcholine is released from the nerve and attaches to receptors on muscle cells. The specific acetylcholine receptors in muscle cells are nicotinic, which is a type of channel protein (7). The receptors respond to the binding of acetylcholine by opening channels that allow sodium ions to pass into the cell, causing depolarization within the sarcolemma (Fig 1). In addition, the action potential opens L-shape calcium channels to bring in calcium from the extracellular fluid. The depolarizing signal travels down T tubules to reach the sarcoplasmic reticulum of the cell. The sarcoplasmic reticulum contains big stores of calcium to be used for contraction (6). Specific ryanodine receptors (RyRs) found on the surface of the sarcoplasmic reticulum are stimulated by the depolarization, and the receptors conformationally change to elicit a mass release of calcium. The high amounts of calcium now in the cytosol bind to available troponin.



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Figure 1. Skeletal muscle contraction via nerve stimulation. Acetylcholine (ACh) initiates a cellular response by creating a signal that stimulates an influx of cytosolic calcium from the extracellular fluid (by L-type Ca^{2+} channels) and sarcoplasmic reticulum (by RyR). The calcium then stimulates muscular contraction. After the action potential ceases, the SR ion channel pumps (SERCA) open to move excess calcium back into the SR. The lack of calcium causes muscle relaxation.

From *Signaling in Muscle Contraction*, by I.Y. Kuo & B. E. Elrich, 2015, *Cold Spring Harb Perspect Biol* 7(2).
doi: [10.1101/cshperspect.a006023](https://doi.org/10.1101/cshperspect.a006023).

Troponin is an enzyme found in skeletal (and cardiac) muscle cells, and it is activated by calcium. Once calcium binds, troponin changes in structural conformation to stimulate tropomyosin. Tropomyosin, another enzyme, binds to the actin-myosin crossbridge binding sites (on the myosin head) during muscle relaxation (6,7). It regulates the muscle fiber to ensure contraction does not happen during rest. These two enzymes are found complexed together (Fig.

2, A). When contraction is signaled, the troponin (activated by calcium) causes a conformational change to tropomyosin. Tropomyosin then moves deeper in the actin/myosin binding site. This leaves the actin and myosin binding sites open and available to cause constriction. Once the troponin/tropomyosin complexes conform to expose the globular myosin heads, myosin then forms a cross bridge with actin (Fig. 2 B). Once bound, the myosin “walks” along the actin filament, fueled by ATP (8). When ATP binds to myosin, the myosin head conformation changes to break the cross bridge. ATP hydrolysis then alters the freed myosin head structure, causing movement up the actin filament. The myosin head then binds to the next binding site on the actin filament (8). This process continues in a cycle to show a sliding movement of actin and myosin, which accounts for muscle contraction.

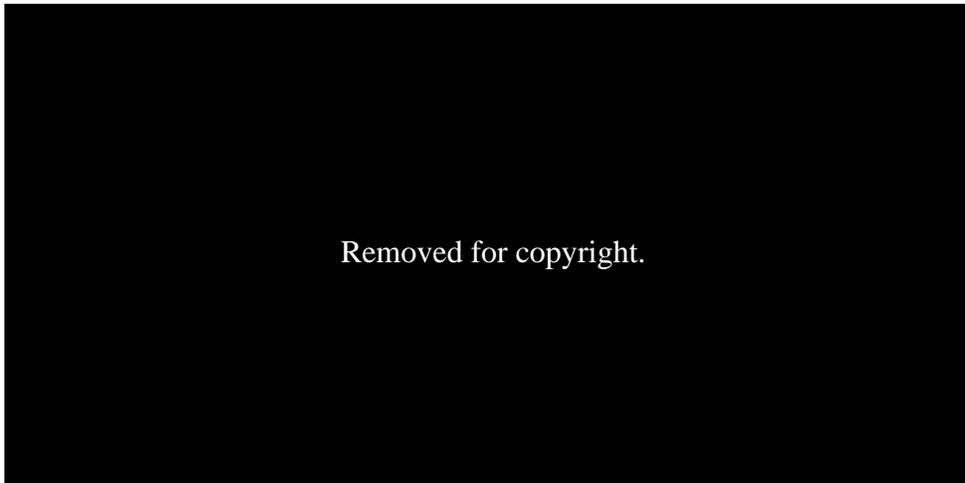


Figure 2. Calcium acts on troponin/tropomyosin to allow for muscular contraction. (A) Actomyosin in skeletal muscle. (1) Relaxed state; tropomyosin/troponin complex is bound to actin/myosin binding sites. (2) Calcium binds/conforms troponin, causing tropomyosin to move furthering the actin groove and therefore revealing the binding site. (B) Actin/Myosin Cross-bridge. (1) Calcium binds to troponin, causing conformational shift in tropomyosin and exposing binding site. (2) ATP binds to the myosin. (3) ATP hydrolysis occurs. (4) The cross-bridge forms and myosin binds to a new position on the actin filament. (5) As the excess Pi leaves, myosin head undergoes conformational change. The myosin head is able to then bind to a new binding site, causing the actin and myosin filaments to slide past each other (overlap). (6) ADP is released, allowing muscle relaxation. From Signaling in Muscle Contraction, by I.Y. Kuo & B. E. Elrich, 2015, *Cold Spring Harb Perspect Biol* 7(2). doi: [10.1101/cshperspect.a006023](https://doi.org/10.1101/cshperspect.a006023).

Muscle relaxation is similarly controlled by calcium. When the nerve signaling contraction halts, repolarization of the sarcolemma occurs. This, in turn, indicates to the sarcoplasmic reticulum to take back up cytosolic calcium. Sarco(endoplasmic reticulum ATPases (SERCAs), which are calcium ion pumps on the sarcoplasmic reticulum membrane, actively transport the calcium in with the help of ATP (7, 8). As calcium leaves the cytosol, troponin is inactivated and unbinds from tropomyosin. Tropomyosin is free then to cover the actin-myosin binding sites. In this way, contraction is inhibited, and actin and myosin return to their unbound state.

Mitochondria

Mitochondria are the main source of energy for most cells. It is especially important in muscle cells because muscle cells require an extraordinary amount of energy for proper contraction. The majority of aerobic respiration takes place within the mitochondria. Cellular respiration takes sugar, mainly glucose, and breaks it down into an energy form, adenosine triphosphate (ATP), that the cell can use for several metabolic reactions (9). The beginning step, glycolysis, takes place in the cytosol of the cell. However, pyruvate, which is the end product of glycolysis, is then moved into the mitochondria. This is where pyruvate is converted to acetyl CoA, which continues through the Krebs cycle. The Krebs cycle happens in the mitochondrial matrix and after several steps, NADH and FADH₂ are generated (9). These molecules then give off their electrons at the inner mitochondrial membrane. The electrons are passed down a series of protein complexes called the electron transport chain, and eventually the electrons power the generation of ATP. ATP is then able to be used by the cell. Contraction of muscle fibers from actin and myosin require a lot of energy, which is why mitochondria take up about 35-40% of the available space in muscle cells (9). Mitochondria also serve to store extra calcium if there is

a high cytoplasmic calcium concentration (10, 11). This is a security measure to ensure that the extremely high amount of calcium ions will not damage muscle cell organelles and proteins.

Myoglobin

Myoglobin is a vitally important component of muscular tissues. It is similar to hemoglobin found in erythrocytes in the fact that they both function to bind oxygen (1). Myoglobin however has a higher affinity for oxygen compared to hemoglobin. In hemoglobin, the oxygen is transported through the blood and then released in different tissues of low oxygen concentrations. In the muscle, the oxygen readily leaves the hemoglobin but is then picked up by the myoglobin molecules (12). The strength of the bond between myoglobin and oxygen is extremely high, meaning that myoglobin is very efficient at storing oxygen in the muscle to use only when absolutely necessary (13). This is essential to efficient cellular respiration, as oxygen is needed for oxidative phosphorylation to produce great amounts of energy quickly. Muscle cells especially need high levels of adenosine triphosphate (ATP) for the actin-myosin action of muscle contraction (9).

Creatine Kinase

Creatine kinase is found in many cells but especially in muscle cells because of the high amount of mitochondria present. It is an enzyme involved in the transport of energy across mitochondrial membranes (to be utilized by myofibrils) via the creatine kinase shuttle (5). Creatine kinase allows a reaction between the highly concentrated ATP and creatine to make phosphocreatine and ADP. In this form, the phosphocreatine can travel through the mitochondrial membrane into the cytosol (14). Once in the cytosol, creatine kinase reverses the reaction to again form ATP and creatine. In this way, energy is effectively shuttled out of the mitochondria to be readily used by the muscle cell.

Sodium and Potassium

Potassium and sodium work together to serve a great purpose in skeletal muscle cells as well. It is seen to be highly involved in cellular signaling, especially in neural cells and neuromuscular junctions. As described earlier, muscular contraction is dependent on the signaling information of the nervous system. More specifically, peripheral nerves carry an action potential from the central nervous system (integrating center) to the neuromuscular junction so that the muscle cells can process the signal and complete the desired action. The signaling is produced by the electrochemical gradients involving potassium and sodium. Nerve cells transport the signal by changing the electric potential of the cell membrane. A normal, polarized cell has a charge of 70 mV at rest. To initiate the signal, sodium channels open on the cellular membrane, and sodium rapidly enters the cell because of passive diffusion. This action changes the cellular potential to about +40 mV. This potential then triggers the adjacent membrane locations to similarly depolarize. This pattern continues down the whole nerve cell until the signal reaches the axon terminal, where neurotransmitters allow synapsis to occur (passing the signal to the next nerve cell). In this way, the signal from the integrating center can efficiently pass information through the peripheral nerves to the muscular cells (via neuromuscular junction).

Repolarization of the cells allow the nerve cells to send future signals to the muscle. This is where potassium plays a big role. When the cell reaches the depolarized potential of about +40 mV, sodium channels close and potassium channels open, causing intracellular potassium to leave the cell via the electrical gradient. This action lowers the cellular potential to about -90 mV, slightly lower than a resting cell. At this point, the electrical potential is close to normal (about -90 mV), but the intracellular levels of potassium and sodium are not at the ideal

concentrations for a repolarized and resting cell. The issue is solved by the sodium-potassium pump, which utilizes active transport to repolarize the neural cells after depolarization occurs (8). The Na⁺/K⁺ ATP pump forces sodium out of the cell and potassium inside of the cell, against their concentration gradients. Potassium, which is a smaller ion than sodium and has open ion channels in the cellular membrane, then leaves the cell according to the concentration gradient to maintain a negative internal environment of -70 mV (repolarization). Depolarization and repolarization are main components of cellular signaling and are especially vital for skeletal muscles because the central nervous system is constantly sending neurological signals to contract and expand muscle groups (15).

It is also observed that exercise of skeletal muscles causes muscle cells to release potassium ions into the extracellular fluid (13). After exercise, potassium concentrations return to normal. The high concentration of potassium in the plasma during exercise serves to further help in exercise efficiency. For example, potassium signals for increased heart rate and ventilation rates that will increase the amount and speed of oxygen entering the body and replenishing muscle tissues. Potassium also initiates vasodilation in the area of the exercising muscle (16). All of these factors together allow for increased blood flow to quickly move oxygen in the tissues and wastes from cellular respiration out of the body.

General Muscle Pathophysiology of Rhabdomyolysis

While different factors contribute to rhabdomyolysis and therefore the initial damage of the muscle cells vary, the general pathophysiology comes from a common biochemical pathway characterized by an initial increase of ionized calcium levels within myocytes (Fig. 3) (10). The increased calcium in muscle fibers activates phospholipase A₂. Phospholipase A₂ is an enzyme that cleaves phospholipids to make fatty acids and glycerol (10, 3). It works by recognizing and

hydrolyzing the sn-2 acyl bond between the second fatty acid chain and the glycerol portion of the phospholipid. Overstimulation of this protein, and other similar calcium-activated proteases such as calpain, cause an abundance of phospholipid breakdown within the muscle cells (15).

The cellular membrane of muscle cells, as well as several organelle, are composed of a phospholipid bilayer. Therefore, an unnatural stimulation of lipid-cleaving enzymes causes degradation of the muscle cell membranes, called the sarcolemma in muscle fibers.

Accumulation of now-free membrane fatty acids and lipids in the cytoplasm causes an upregulation of phospholipases in a damaging circular pattern (10, 17).

The free fatty acids, from membrane breakdown, are closely linked to oxidative stress caused by free radicals (2). Polyunsaturated fatty acids have high amounts of double bonds, which causes the bent shape necessary for proper cellular membrane fluidity. Membranes with high amounts of unsaturated fatty acids also serve to protect against inflammation and oxidizing agents. When these unsaturated fatty acids are interrupted or broken off, the cellular membranes become very vulnerable to damage from free radicals. Free radicals are chemical species that are extremely reactive because of their characteristic unpaired electron. Reactive oxygen species like peroxides, superoxides, and hydroxyls, are a specific form of free radicals that contains an oxygen group and are commonly seen in rhabdomyolysis. These free radicals oxidize molecules (proteins, lipids, nucleic acids, etc.) on vital organelles and cellular components of myocytes (2, 15). Oxidizing these molecules instigate conformation changes that lead to functional issues.

The free fatty acids, hydrolyzed by free radicals, especially effect the nucleic acids of the cell's DNA (deoxyribonucleic acid). Chemical interactions with the DNA cause genetic mutations that lead to abnormal enzyme transcriptions and diminished cellular function. In the case of mitochondrial DNA mutations, the transcription of enzymes involved in cellular respiration,

more specifically oxidative phosphorylation, is inhibited (18). This is one source of ATP depletion of the muscle cells.

Reactive Oxygen Species also affect important muscular organelles including the sarcoplasmic reticulum. They inhibit the original structure of the organelles, causing a decrease or complete inhibition of normal biological function. Because the sarcoplasmic reticulum (SR) plays such a big role in calcium storage for muscle contraction, destruction of SR membrane lipids and carrier proteins results in a high concentration of cytoplasmic calcium (10, 18). Naturally, the calcium binds to the troponin molecules. The now-activated troponin changes chemical conformation to pull tropomyosin off of the actin-myosin cross-bridge binding sites of the muscle fiber. This in turn allows the normal actin-myosin binding of the muscle fiber. However, in the case of too much calcium and other oxidative chemicals, the actin-myosin cross bridging cannot be stopped. Too much calcium causes prolonged troponin and tropomyosin binding, and the myosin binding sites on the actin stay available far too long for normal contraction (3). The continuous contraction of skeletal muscles expends a lot of energy. Because each attachment of a myosin head to an actin binding site costs an adenosine triphosphate molecule (ATP), persistent contraction causes a rapid depletion of ATP reserves. Muscle cells consume a lot of bodily energy within normal pathology, so this condition causes a detrimental energy gap. This can lead to compensating biological pathways like excessive purine metabolism (10). Calcium buildup inside the plasma membrane also leads to an influx of water inside the cell along with unwanted activation of different proteins (18). As the cell's integrity is greatly disturbed, the influx of water further breaks apart the sarcolemma. Cell inflammatory responses are activated, and the combination of excess calcium, water and ATP depletion leads to the rapid deterioration of muscle cells.

Increased cytosolic ionized calcium in muscle fibers also leads to an increase in mitochondrial calcium. The abnormally high concentration of calcium of the cytoplasm compared to the lower calcium levels of the mitochondria instigate passive transport into the mitochondria (11). Mitochondria have naturally high amounts of calcium, so an unnatural influx of calcium leads to complications. Prolonged influx of calcium effects the structural integrity of mitochondrion, leading to functional issues. Oxidative phosphorylation, which occurs in the mitochondrial inner membrane, accounts for the vast majority of ATP production for the body (11). Therefore, if this process is impeded by the excess calcium, energy production is significantly slowed. Starving cells of ATP, especially skeletal muscle cells, inhibits several necessary cellular reactions. Among the effected chemical reactions are the vital calcium carrier proteins and ion channels of the sarcolemma. This repeats the detrimental cycle of increased cytosolic calcium and organelle destruction (17). On a widespread scale, this cycle can quickly amount to serious damage and even necrosis of muscle tissues.

Unhealthy increase in calcium also activates cytochrome c and AIF. Cytochrome c is a heme protein located along the electron transport chain, found within the inner mitochondrial membrane (11). It plays an integral part of the oxidative phosphorylation process by transporting electrons from enzyme cytochrome c – oxidoreductase (complex III) to enzyme cytochrome c oxidase (complex IV) (10, 19). However, when cytochrome c is found free within the cell's cytoplasm, it signals the start of cellular apoptosis. Freed cytochrome c is the product of mitochondrial ROS damage, as described previously. Cardiolipin, a lipid that anchors cytochrome c to the mitochondrial membrane, is attacked by ROS at its hydrophobic tail (2). The cytochrome c is then free to pass through the pores of the outer mitochondrial membrane and accumulate within the muscle cell. Cytochrome c activates the protease caspase 9 (18). This

leads down the pathway to intrinsic apoptosis. AIF, apoptosis inducing factor, is another protein that is upregulated by high levels of calcium found both in the nucleus and in the mitochondria (10). It works to stimulate the condensation of chromatin and nucleic elements so that DNA is broken down. This is in preparation of cellular death.

All of these damaging processes contribute in myocyte destruction. Organelles break down and the sarcolemma deteriorates, resulting in the release of cytosolic chemicals into the interstitial fluid surrounding the muscle tissues. When this process affects multiple cells, the concentrations of toxic and oxidized molecules and excess cellular fluid increase to unhealthy levels (5). This causes adverse effects on otherwise healthy neighboring cells. Buildup of fluid causes localized edema. The pressure of lysed muscular cell contents also inhibits blood, and therefore nutrient and oxygen, flow to neighboring tissues. Ischemia and muscular atrophy follow. The concentration gradient of the toxins leads to the diffusion of harmful chemicals throughout the surrounding tissues and into the bloodstream (10). This is where the chemicals can cause widespread damage not only to skeletal muscle, but also to the rest of the body. Creatine phosphokinase (CPK), ionized calcium, BUN/creatinine, potassium, and uric acid are among the unhealthy molecules released from myocytes (1, 4). In the natural settings, these molecules and proteins are useful, and even essential, to the body. However, when put in the wrong physiological environment, these substances lead to harmful results.

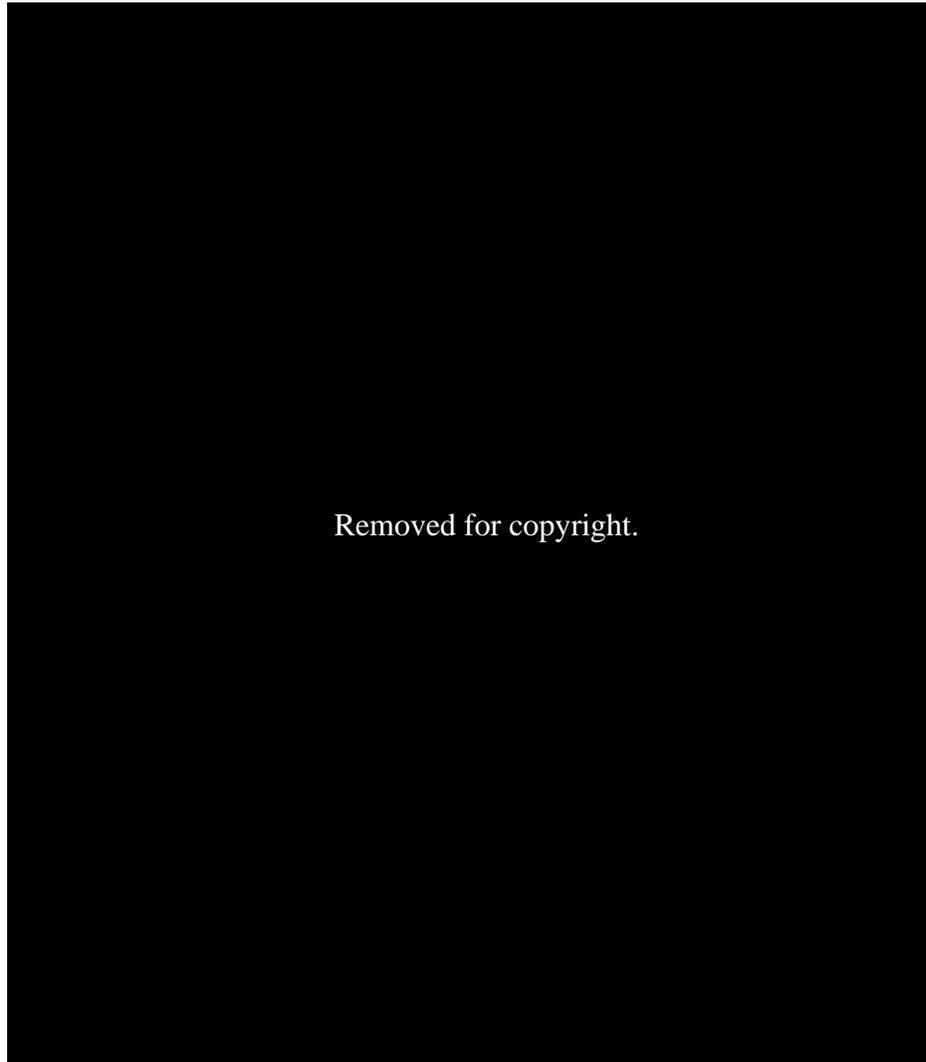


Figure 3. Pathophysiological mechanisms of rhabdomyolysis. Decreased ATP supply and sarcolemma rupture increase levels of free ionized calcium, causing various intracellular pathways/cascades that result in muscle tissue breakdown. Intracellular substances become toxic as they leave the damaged muscles and disperse through the body. $[Ca^{2+}]_c$: cytoplasmic Ca^{2+} , $[Ca^{2+}]_m$: mitochondrial Ca^{2+} , ROS: reactive oxygen species, PLA2: phospholipase A2, - - -: feedback. From *The syndrome of rhabdomyolysis: Pathophysiology and diagnosis*, by Giannoglou, G. D., Chatzizisis, Y. S., & Misirli, G., 2007, *European Journal of Internal Medicine*, 18(2). doi:10.1016/j.ejim.2006.09.020

Causes of Rhabdomyolysis

Rhabdomyolysis is a multifaceted condition because it can be triggered by a number of different causes. In addition, several smaller, and sometimes unrelated, factors can combine to contribute to the rapid break down of muscle tissue. A lot of times, this makes locating a clear cause challenging for medical professionals. Several causes have been recognized to directly contribute to rhabdomyolysis. The causes are generally broken into two categories: traumatic and non-traumatic. The most common cause is direct muscular trauma, such as blunt-force trauma or a crush injury. Prolonged exercise, severe burns, ischemia, and prolonged muscle immobilization can also contribute to traumatic rhabdomyolysis (13,15). There are also many non-traumatic contributors that can cause rhabdomyolysis. This includes drug abuse, snake and spider bites (venom), bacterial infections, malignant hyperthermia, heat stroke, and metabolic/genetic disorders (20, 21).

Traumatic Rhabdomyolysis

Crush injury and blunt force trauma. Crush injury itself is a broad term to describe bodily harm. Crush injury normally occurs when a body is squeezed or pinched between two heavy objects (18). This can manifest from collapsed buildings, car accidents, earthquakes, and other natural disasters. Blunt force trauma affects the body similarly. This generally occurs when something with great mass or with strong force collides with a body. This can happen when a person is hit by a car, a baseball bat, rubble during a bombing, or even impact from another person's fist. These types of trauma can cause great force on the body, especially on the internal organs and muscle groups. As the force from the object presses against the skeletal muscle, the muscle fibers rub against each other and the sarcolemma begin to break apart (3). The membranes of organelles within the body, such as mitochondria and sarcoplasmic reticulum,

also disintegrate, releasing their contents. These muscle fiber contents leak into the surrounding interstitial fluid. At the same time, nearby blood vessels rupture under the force (5). This causes uncontrolled hemorrhage that pools in the located muscle area. The blood and myocyte cellular contents, within the interstitial fluids, mix. When the pressure of the object crushing the victim is removed, the fluid and blood are released. The muscular chemicals, including myoglobin, creatine phosphokinase, calcium, and potassium, displace through the circular system and diffuse throughout the rest of the body (10). In serious crush injuries and blunt force trauma, muscular necrosis, myoglobinuria, and hypovolemic shock ensue (18). These are potentially life-threatening conditions. Milder crush injuries can cause swelling and compartment syndrome.

Burns. In addition to blunt force trauma and crush injury, muscle injury due to burns can cause rhabdomyolysis. Third degree burns from fires, lightning strikes, and other forms of electrocutions damage muscle tissue directly (22). It has been observed that up to 10% of electrical burn patients develop rhabdomyolysis (2). This form of extreme damage of the muscle and exposure of muscle to the external environment leads to necrosis. Inflammation of the affected area ensues, and prolonged immune response leads to chronic adrenergic stress. Again, the result of this form of trauma is rhabdomyolysis, with the spreading of unwanted myocyte contents.

Over-exertional exercise. Exercise induced rhabdomyolysis has been observed within athletes and non-athletes alike. Even the most elite athletes, like professional football players and endurance runners, have fallen victim to rapid muscle breakdown (15). This form of rhabdomyolysis is thought to be underreported somewhat due to a lack of the typical symptoms (brown urine, muscle pain, fatigue, etc.). Although seen throughout different types of sports and aerobic activities, the common factor of this form of muscle breakdown is too much exercise

during too long of a time period. Examples of highly strenuous forms of exercise that can increase the chance of rhabdomyolysis include prolonged heavy weightlifting and marathon running. Although the exact biochemical route of muscle breakdown varies from athlete to athlete, it is thought that the high body temperature, dehydration, and electrolyte imbalance all contribute (23).

Naturally, a body's temperature increases during muscle movement. The body responds to the change of temperature by sweating and dilating blood vessels. Extreme sweating can lead to the rapid loss of salts (sodium, chloride, magnesium, calcium, etc.) and water (15). Rapid loss of water and salt effects the proper functionality of myocytes. The ability for the sarcoplasmic reticulum to release calcium and activate sarcomeres is diminished. Lack of water causes myocyte crenation which damages its organelles and molecular contents. Hot body temperature can also directly damage muscle cells by creating a hypermetabolic state (17). The bonds of muscle cell lipid bilayers begin to break and cytoplasmic materials spill into interstitial fluid. This version of rhabdomyolysis can be at higher risk for those diagnosed with sickle cell and hypokalemia (23). In addition, extreme exercise can also lead to physical tearing of muscles. Common presentations of muscle injuries include hamstring, quadriacep, and calf muscle tears (17). This version of muscle injury acts like a crush or blunt impact injury in that muscle tissue is physically broken apart. The fascia that holds fasciculi together rip apart under force, and the muscle fibers further breakdown until the cell membrane loses integrity (15, 17). Much like the other forms of rhabdomyolysis, muscular chemicals spread throughout the circulatory system and lead to bodily harm.

Prolonged immobilization. Seemingly opposite of over-exercise, prolonged immobilization of a body also leads to rapid muscle breakdown. This type of muscle injury presents similarly to a crush injury, as muscle tissue is compressed. Generally, when the body is immobilized, the weight of the body's skeleton, internal organs, and other muscle groups is enough to place heavy pressure on certain muscles (12). Normal body rest due to sitting and sleeping is not enough to cause this type of rhabdomyolysis, however. For there to be noticeable muscle breakdown, the immobilization must be prolonged. Instances of persistent immobilization include seizures, alcoholic-induced unconsciousness, and anesthesia (21). These conditions cause a person to lose consciousness at unpredictable moments, and he or she is prone to collapsing. Muscle breakdown is further agitated if the body collapses on a hard surface. As the body lays on the floor, myocytes begin to break down mechanically and chemically. Mechanical breakdown occurs by the force of the body pressing on the muscle. In addition, arterial vessel walls are being compressed. This leads to greatly diminished blood flow to the effected muscle tissues (17). Consequently, the myocytes begin to chemically starve from lack of nutrients and oxygen. Trapped blood and interstitial fluid swell and myocyte contents leak out. Like crush trauma, the most harmful part of the injury, in regard to muscle breakdown, occurs when the body is moved. This is because the great pressure that retained the toxin-filled interstitial fluid is released, and therefore the muscle cell components propel throughout the circulatory system.

Non-traumatic Causes

Drugs: prescription and elicit. Numerous types medications have the possibility of causing rhabdomyolysis. A very common type of drug to induce skeletal muscle illnesses are statins. Statins are HMG-CoA reductase inhibitors, which lower cholesterol levels (4, 10).

Statins are usually prescribed when a patient is at risk of cardiovascular disease because of plaque buildup. Muscle impairment due to HMG-CoA reductase inhibitors occurs in about 0.1% cases, but because statins are a commonly prescribed medicine, there is concern about the increase of rhabdomyolysis occurrence (24). By inhibiting HMG-CoA, statins stop the bodily production of mevalonate. Mevalonate is essential precursor of cholesterol, which is why statins help lower the risk of heart disease (17). However, mevalonate is also needed to make isoprenoids, which are lipids that help anchor GTPases to cellular membranes. GTPases have many roles within the cell, including cytoskeleton formation, apoptosis regulation, and internal cell signaling (24). Inhibiting GTPase attachment to the lipid membranes therefore compromises the integrity of muscular cells. This is where cell membranes disintegrate and cellular contents spill into other areas of the body.

In addition to statins, many illicit drugs like methamphetamine and certain opioids have been observed to induce rhabdomyolysis. Generally, these drugs cause extreme agitation of muscular tissue by sarcolemma irritation and protein inhibition (25). Cocaine is another drug that has caused rapid muscle breakdown. Cocaine increases stimulation within the body by binding to dopamine transporters, which block the removal of dopamine from the synapse. This stops norepinephrine re-uptake in the nervous system and amplifies the signals (3, 26). This overstimulation from cocaine can lead to hyperactive muscle activity (at deleterious levels), widespread ischemia-causing vasoconstriction, and drug induced unconsciousness. All of these conditions contribute to rhabdomyolysis by either mechanical or chemical means. A lot of times, a combination of medications can cause adverse effects within muscle, leading to acute intoxication or comatose state.

Venom. Another non-traumatic cause of rhabdomyolysis is a bite from a venomous animal. These types of bite are commonly found to occur with certain species of snakes, including the North American copperhead (*Agkistrodon contortrix*) and timber rattlesnakes (*Crotalus horridus*) (26). Many of these venomous snakes have hollowed fangs that first poke into its prey and then glands secrete the venom through the fang, directly into the prey. Certain species of spiders, like the brown recluse (*Loxosceles reclusa*) and western black widow (*Latrodectus variolus*) also have a venomous bite. These are among the most common venomous animals in Northern America. Similar to snakes, spiders also have small fangs that protract into its prey and deliver the venom. While each species produces slightly different versions, the venom of these animals all contains proteins and polypeptides designed for killing prey and protection (26). The main goal of the venom is to immobilize and digest tissue, so most animal venoms contain phospholipases. Phospholipases are enzymes that aid in the breakdown of phospholipids. Phospholipase A₂ (PLA₂) especially is seen to be one of the main components of venom that contributes to its toxicity (10). PLA₂ of venom breaks down phospholipids involved in the blood clotting cascade process, which causes unhealthy prolonged blood coagulation (26). The vessels around the injection site are physically damaged from the bite. The blood gels, which inhibits oxygen transport to surrounding muscular tissue. Ischemia leads to cell death, as the cell is unable to generate energy. In addition, the phospholipase A₂ also directly affects muscle cells. As described earlier, muscle cell membranes, and many organelles, are made up by a phospholipid bilayer. When PLA₂ is injected into muscular tissue, the components of the membranes themselves are hydrolyzed, disrupting membrane structure and spilling the intracellular contents into the extracellular space (10, 17).

Complications of Rhabdomyolysis

The effects of rhabdomyolysis range from undetectable conditions to minor illnesses to lethal conditions, depending on the severity of muscle damage and death. The reason rhabdomyolysis can be so concerning is because so many complications can result. Some of the major complications include acute renal failure, compartment syndrome, hyperkalemia, hypocalcemia, and hepatic inflammation (Fig. 4). These complications are widespread and dependent on the severity of the muscle breakdown.

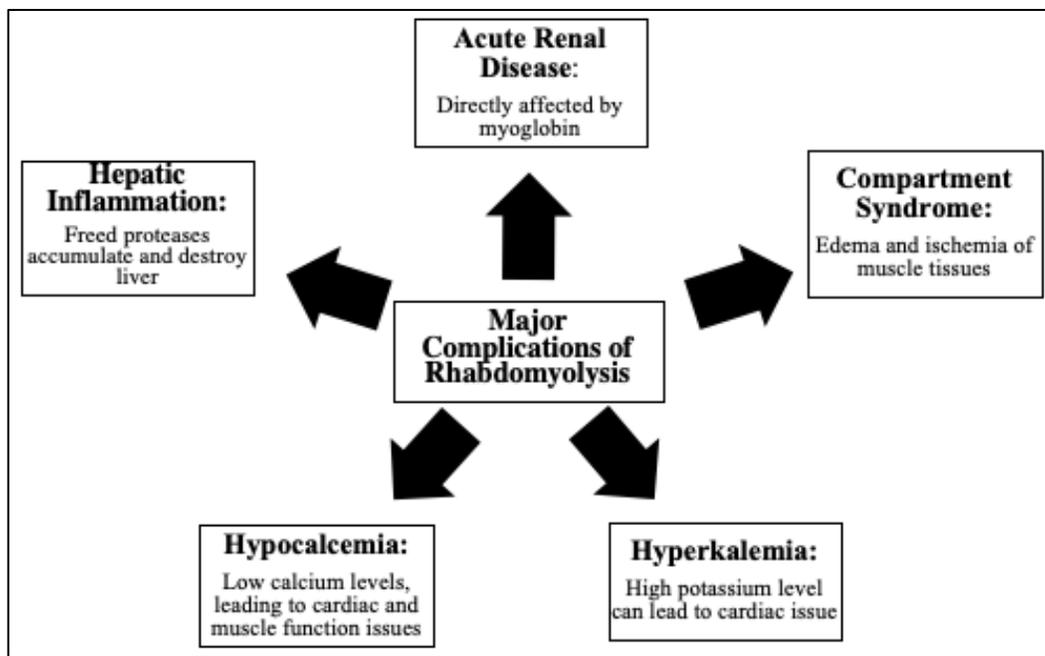


Figure 4. The Major Complications of Rhabdomyolysis.

Acute Renal Disease

One of the most severe complications of rhabdomyolysis is renal disease and failure. In fact, about 33% of all reported cases of rhabdomyolysis in the United States are shown to develop acute renal injury (2). This complication is potentially lethal. Muscle cell breakdown leads to kidney problems because of the many proteins and compounds that are released into the circulatory system. This is usually detected by the classic symptom of dark-colored urine (13).

Myoglobin is the main contributing molecule of acute kidney injury. Although myoglobin is very useful within muscle fibers, it can be damaging when misplaced in the body. Myoglobin floods the bloodstream during rhabdomyolysis, and this blood is eventually brought to the kidneys. The kidneys main function is blood filtration for toxin excretion. Therefore, when the unwanted muscular cell contents flow through the body, the kidney works to get rid of the potentially toxic elements (10, 13).

Nephrons are the functioning unit of the kidney, and they function on a microscopic scale. The four main functions of nephrons are fluid filtration, secretion, absorption, and excretion. The filtering portion of the nephron is called the glomerulus (27). The nephron also has proximal and distal convoluted tubules and the loop of Henle, which aid in electrolyte reabsorption and secretion. The collecting duct of the nephron then allows the filtrate to be sent to the bladder and eventually excreted from the body. Myoglobin, along with the other blood contents, is brought to the kidney via the renal artery. Renal vasoconstriction is the first complication from myoglobin, as indicated by Figure 5 (3). Because of myoglobin's high affinity for oxygen, it has a tendency to bind to other oxygen-containing molecules, especially nitrous oxide. Nitrous oxide is a vasodilator and plays a pertinent role in the regulation of renal hemodynamics. If excess myoglobin binds to nitrous oxide, the nitrous oxide cannot be used to dilate the renal arteries and veins (1, 3). Continuous constriction of the renal artery leads to hypertension and a decreased glomerular filtration rate, which drastically reduces kidney function.

Once the myoglobin arrives at the glomerulus, it is able to pass through the filtration system (glomerular basement membrane) and enters the nephron's proximal and distal tubules. Here, myoglobin concentrates within the filtrate. Acidic urine within the nephron creates a

reactive environment for the myoglobin to readily react with Tamm-Horsfall, or uromodulin, protein (3). Uromodulin is the most common protein excreted in urine and serves to prevent calcium calculi (kidney stone) formation (13). However, in acidic environments, uromodulin and myoglobin precipitate within the tubules of the kidney. Buildup of this precipitate, called intratubular casts, causes nephrotic obstruction and is usually observed within the distal tubule (1). Rhabdomyolysis also triggers dehydration, so with less water present than normal, these intratubular casts cannot easily be broken down or dissolved.

Lastly, myoglobin can directly inhibit nephrotic tubular cell function, especially if high concentrations build up in partially obstructed nephrons. Because myoglobin is a monomer protein containing a heme group, myoglobin is very reactive towards oxygen. If the heme breaks away from the myoglobin molecule, it can cause great oxidative stress. The renal epithelial cells lining the tubules of the nephron are then exposed to damage from the oxidative heme group (3). Certain phospholipids of the cellular membranes are pulled away by the heme groups, destroying the integrity and sometimes killing the cells uncontrollably. The end result is acute tubular necrosis. When a vast amount of muscle is damaged, severe kidney damage due to tubular obstructions, vascular constrictions, and oxidative stress can lead to kidney failure (13). Kidney failure can be extremely lethal, especially if both kidneys are being affected by rhabdomyolysis.



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Figure 5. Mechanism of Acute Renal Failure caused by rhabdomyolysis. Muscle tissue breakdown releases proteins toxic to the kidneys. Dehydration and intestinal mesenteric ischemia occur in response to intense systemic vasoconstriction (caused by myoglobin binding to Nitric Oxide). Hazardous tubular casts form and myoglobin further damages nephrotic tissue via its radical properties. All of this combines to extensively damage the kidneys. From Rhabdomyolysis updated, by Efstratiadis G. & et al., 2007, . Hippokratia 11(3):129–137. PMID: 19582207

Compartment Syndrome

In addition to renal injury, there are other conditions that arise as a result of rhabdomyolysis, including compartment syndrome. This generally occurs after muscle trauma or ischemic conditions, where a group of damaged muscle tissue causes severe hypertension

within its immediate environment/tissue compartment (18). The high pressure causes a lack in blood flow and induces further ischemia and a lack of nutrient flow, therefore triggering additional damage to the muscles. Eventually, the pressure rises enough (>30 mmHg) to compromise surrounding arterioles, and fluid flow to the area completely stops (2). The result of this is necrosis of the infected area. If left untreated, compartment syndrome can permanently damage a muscle group or even an entire limb.

Hyperkalemia

Hyperkalemia is another result of rhabdomyolysis. Hyperkalemia means the levels of potassium in the blood are too high. Potassium levels within the 3.6 to 5.2 mmol/L range are expected within normal physiology (12). However, rapid muscle breakdown can increase the concentration of potassium to unhealthy levels. Because potassium plays a big role in muscle and nerve signaling, high potassium levels can instigate cardiac arrhythmia and even cardiac arrest (5). Hyperkalemia effects the electrical activity of cardiac muscle responsible for normal heart contraction and relaxation. In serious cases, the high potassium can completely inhibit electrical signaling of the heart. This radical version of hyperkalemia is fatal, while less severe versions lead to less drastic illness including kidney stone formations and glomerular nephritis.

Hypocalcemia

Because calcium is so involved with the molecular pathophysiology, hypocalcemia can result. Hypocalcemia is the condition of low blood plasma levels of calcium. Where the normal range of calcium in the blood is about 2.1-2.6 mmol/L, some rhabdomyolysis patients have lower numbers (12). Release of phosphates during muscle breakdown cause calcium phosphate to form within bodily fluid, which pulls away free calcium ions necessary for bodily function (10). Low calcium levels can affect cardiac muscle function, similar to hyperkalemia. Mild

hypocalcemia can be asymptomatic, but severe conditions (perhaps from extreme rhabdomyolysis) lead to heart arrhythmia, muscle spasms, and sometimes seizures (1).

Hepatic Inflammation

Hepatic inflammation can be seen in about 25% of patients diagnosed with rhabdomyolysis (5). This complication involves the inflammation of the liver due to damage by intracellular myocyte contents. While inflammation and damage of the liver is most likely caused by many of the cytosolic enzymes, it is especially observed that freed proteases attack liver tissue (28). During rhabdomyolysis, these proteases escape their cells and are transported through the circulatory system. The liver is designed to filter the circulating blood, so eventually the high concentration of myocyte proteases congregates in the liver. As the liver attempts to detoxify the blood, the proteases function to break down hepatic cells. This leads to the immune response of inflammation. Liver inflammation is serious because low liver function leads to the retention of toxins within the circulatory system (5). This can lead to a multitude of health issues.

Diagnostics of Rhabdomyolysis

Because there are varying causes of rhabdomyolysis, it is important to gather adequate information about a patient's condition and history to properly diagnose this condition. In addition, it is very important to closely analyze and monitor the symptoms of a possible rhabdomyolysis patient. With crush victims, there is a high chance of rhabdomyolysis. Listening for complaints of muscle spasms, pain, or aching in skeletal muscle areas is key to catching this condition before severe consequences occur (12). With non-traumatic patients, there may be an absence of typical symptoms, so it is especially important to recognize the symptoms. The typical three symptoms that accompany rhabdomyolysis are reddish-brown

urine, muscle pain, and muscle weakness. In most cases, the location of muscle pain is the same area where rhabdomyolysis is occurring. Commonly, rhabdomyolysis patients complain of pain in thigh and shoulder areas (5). Muscle pain is not always detected with rhabdomyolysis, however. Occasionally, seizures, swelling, and muscle spasms are seen (4). The variability of severity and symptom presentation makes this disease important to diagnose through testing. There are quite a few different ways clinicians can confirm a rhabdomyolysis diagnosis in addition to observing a patient's symptoms. Easy tests that can be performed include urine and blood tests. These measure levels of myoglobin, creatine phosphokinase, certain electrolytes, and BUN/creatinine in the blood (12). A forearm exercise test can also be performed to further investigate muscle damage, as abnormally low lactate levels during exercise indicates metabolic issues associated with rhabdomyolysis.

Urine Dipstick Evaluation

The components of urine are very important in diagnosing rhabdomyolysis. Not only does the color of urine provide clues to what is happening inside a body, but urinalysis can confirm irregular chemical levels. A urine dipstick test can test for acidity, glucose levels, proteins, blood cells, and bilirubin (10). Myoglobin presents as a reddish-brown protein and is responsible for the characteristic dark urine of most rhabdomyolysis cases. For myoglobin to be visually identified in urine, there must be a concentration of about 100mg/dl (10). However, diagnostic testing of myoglobin in urine or serum is much more specific. If a dipstick test performed on a urine sample comes back positive for blood but with no indication of erythrocytes, myoglobinuria is likely (12). Testing for potassium and calcium levels through a urine sample is also important for diagnosing rhabdomyolysis. It is especially important to test

for potassium and calcium levels since an excess or lack of these ions can lead to heart and epileptic complications.

Blood Serum Sample

Blood serum testing is also useful in identifying rhabdomyolysis. This form of testing can indicate if BUN/creatinine levels are abnormal. BUN stands for Blood Urea Nitrogen, so this test measures how much nitrogen is present in a person's blood (5). Muscles cells contain large amounts of proteins, and therefore a lot of amino acids. Amino acids are made up of a carboxylic acid group (-COOH), an R group that varies, and an amine group (-NH₃). When muscle breakdown ensues, the amino acids are broken down so that the amine groups (urea) are freed. Liver processing further breaks down urea, resulting in the formation of excess nitrogen in the blood (28). Therefore, if rhabdomyolysis is suspected, one would look for an increase in blood nitrogen levels via a (BUN) test. Likewise, creatinine is also formed when muscle cell metabolism occurs. Creatine phosphate donates its phosphate to adenosine diphosphate to make adenosine triphosphate (ATP) (10). The waste of this reaction is creatinine. In the event of rhabdomyolysis, muscle tissues are under stress. Whether from extreme exercise, ischemia, or a crush injury, the muscle cells are in dire need of nutrients and energy. To compensate, the cells use up all available energy that is stored in creatine phosphate. This leads to very high levels of creatinine. Measuring creatinine concentrations in blood can help confirm a rhabdomyolysis diagnosis (5).

Another easy diagnostic test to perform when suspicion of rhabdomyolysis arises is an anion gap test. An anion gap test identifies common electrolytes of the body, including sodium, potassium, chloride, and bicarbonate. This test is also determined through a blood sample. By looking at the anion gap levels, the pH of a patient's blood can be determined. Rhabdomyolysis

generally can create an acidic environment (10). Therefore, if an anion gap test indicates metabolic acidosis, rapid muscle breakdown may be likely. Measuring serum lactate levels is another way to confirm rhabdomyolysis. Lactic acid is an organic acid product of anaerobic respiration (5). Cellular respiration is the normal way a cell harnesses energy from nutrients. However, cellular respiration with the Krebs cycle and electron transport chain is an aerobic process. This means that it requires oxygen to work. When oxygen is not readily available, the cell can perform anaerobic respiration (15). Glycolysis occurs so that pyruvate is made from glucose. Then, a protein called lactate dehydrogenase converts the pyruvate to lactic acid. This produces a small yield of ATP, although it is not as efficient as aerobic respiration. Lactic acid production occurs within normal physiology during exercise, when oxygen is scarce. However, rhabdomyolysis can also display high lactate levels (5, 15). Ischemia and traumatic crush injury can limit oxygen supply greatly. Therefore, high lactic acid levels can help further confirm a rhabdomyolysis diagnosis.

Forearm Exercise Test

In some instances, rhabdomyolysis can result from certain inherited predispositions. Disorders in metabolic processes, such as glycogen metabolism, oxidative phosphorylation, and fatty acid beta-oxidation, can greatly raise the chance of developing rhabdomyolysis. This makes sense, as these processes deal with the ability of breaking down molecules for quick energy availability. Muscle tissue is detrimentally affected by these forms of metabolic disorders, because the process of muscle contraction requires a great deal of ATP. Because rhabdomyolysis is a likely result of these glycogen storage disorders, a forearm exercise test can be performed to help diagnose these predispositions. This is an indirect way to preemptively stop rapid muscle breakdown (or at least make the patient aware of their increased likelihood of

forming rhabdomyolysis). This test works to identify the changes in lactate, pyruvate, and ammonia levels during exercise (5). A patient's baseline levels are measured via a catheter in a vein of the forearm (12). After a baseline test, a blood pressure cuff is utilized, and the patient squeezes an ergometer for a minute. The blood pressure cuff is deflated, and another blood sample is immediately collected. Samples are also taken at 3, 5, 6, 10, and 15 minutes (12). Healthy individuals should display an initial spike of lactic acid and then a gradual decrease to normal physiological levels, indicating normally functioning metabolic pathways. However, a forearm exercise test displaying high lactate and ammonia levels indicates a metabolic issue. An inadequate rise of lactate paired with a normal rise in ammonia suggests an issue of glycogen breakdown to pyruvate (glycogen metabolism disorder). A result containing abnormally low lactate levels and an elevated level of pyruvate implies a deficiency with the enzyme lactate dehydrogenase. These issues are known to prompt rhabdomyolysis, so identifying the metabolic issues during times of muscular exercise allows the patient to make better informed choices regarding health and exercise. Those predisposed to certain metabolic issues may need to adjust their activity/exercise habits in order to maintain healthy muscle tissues.

Generally, when considering rhabdomyolysis, one must look at all of the evidence. Urine and blood levels are very useful in confirming muscle breakdown when used in conjunction with other symptoms. It is important to listen to patient complaints of muscle pain and weakness as well as assessing any form of injury or background information (10). Especially in instances of obvious crush injuries, new medications, and prolonged exercise, rhabdomyolysis should be suspected and confirmed through diagnostic testing.

Management and Prognosis of Rhabdomyolysis

As of now, the main management method for rhabdomyolysis is an early diagnosis paired with the quick removal of the harmful muscular molecules (myoglobin, electrolytes, proteins, etc.). In fact, one might argue that the quickness of treatment is the main determining factor of survival and severity of adverse effects for this certain condition. Of course, the primary action would be to stop whatever was causing the muscle necrosis, whether this means carefully removing a heavy object with traumatic rhabdomyolysis or discontinuing a certain medicine with non-traumatic rhabdomyolysis. After the initial cause of rhabdomyolysis has been determined, the condition can be treated through several standard methods. One of the main managements of rhabdomyolysis is intravenous hydration. Most commonly, a saline solution (NaCl 0.9%) is inserted via IV needle into a forearm (3). It is important to supply the circulatory system with excess fluids to first dilute the toxic levels of the myocyte intracellular contents. Adding fluids also serves to flush the toxic molecules away from the muscle tissue and allows for proper detoxification via kidneys and liver. Increased fluid content also increases glomerular filtration rate of the nephron, which improves kidney function (29).

Another affective way of managing rhabdomyolysis is by increasing bodily pH. Bicarbonate and mannitol can be administered to a patient to stimulate urine alkylation (2). The especially acidic environment of nephron tubules induces myoglobin damage. Therefore, increasing the pH via bicarbonate flushes out the concentrated myoglobin from the tubules. In addition, mannitol helps increase blood flow by vascular dilation which also helps flush toxins from the kidneys (13). Other treatment methods include calcium supplementation and potassium reduction methods, but these are secondary compared to the treatments focused on kidney function (1). Of course, dialysis is a drastic management method for the most severe cases of

rhabdomyolysis. Dialysis is the process where water, toxins, and excess fluids are mechanically removed from blood. It is generally only used in patients with severe kidney damage because the process is extensive. A patient has his blood taken out of his body and run through various membranes to sort out certain molecules. In the case of rhabdomyolysis, myoglobin, BUN/creatinine, lactate, and excess potassium compounds would be among the chemicals to be filtered out (13). The filtered blood is then returned to the patient. Dialysis has shown to be effective in treating rhabdomyolysis.

Rhabdomyolysis comes in a variety of forms, from extremely mild to possibly fatal. Generally, the prognosis is fairly good if renal failure is not detected. Pre-existing conditions and a culmination of physiological factors contribute to severity. However, when diagnosed early and treated punctually, there is a very good chance of full recovery. It was estimated that rhabdomyolysis (without acute renal injury) has a mortality rate of 8-10% (5). When the kidneys were damaged during rhabdomyolysis, though, the mortality rate increased to 51% (5). Because rhabdomyolysis can form from a diversity of causes, it is important for prompt medical diagnosis via background information, symptom presentation, and simple urine/blood testing. A quick diagnosis ensures minimal complications and greatly increases the prognosis.

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