Hylonome

Review Article

The creatine kinase response to resistance exercise

A.J. Koch[1], R. Pereira[4], M. Machado[2,3]

[1]Exercise Physiology Laboratory, Lenoir-Rhyne University, Hickory, NC USA; [2]Laboratory of Human Movement Studies,
Universitary Foundation of Itaperuna (FUNITA), Itaperuna, RJ, Brazil; [3]Laboratory of Physiology and Biokinetic,
Faculty of Biological Sciences and Health, Iguaçu University Campus V at Itaperuna, RJ, Brazil;
[4]Department of Biological Sciences, State University of Southwest Bahia (UESB), Jequie, BA, Brazil

Abstract

Resistance exercise can result in localized damage to muscle tissue. This damage may be observed in sarcolemma, basal lamina, as well as, in the contractile elements and the cytoskeleton. Usually the damage is accompanied by release of enzymes such as creatine kinase (CK) and lactate dehydrogenase, myoglobin and other proteins into the blood. Serum CK has been proposed as one of the best indirect indicators of muscle damage due to its ease of identification and the relatively low cost of assays to quantify it. Thus, CK has been used as an indicator of the training intensity and a diagnostic marker of overtraining. However, some issues complicate CK's use in this manner. There is great interindividual variability in serum CK, which complicates the assignment of reliable reference values for athletes. Furthermore, factors such as training level, muscle groups involved, and gender can influence CK levels to a greater extent than differences in exercise volume completed. This review will detail the process by which resistance exercise induces a rise in circulating CK, illuminate the various factors that affect the CK response to resistance exercise, and discuss the relative usefulness of CK as a marker of training status, in light of these factors.

Keywords: Enzymes, Muscle Damage, Weight Training

Introduction

Resistance exercise can result in localized damage to muscle tissue. Eccentric muscle actions are most often implicated in etiology of skeletal muscle damage, as they consistently produce the greatest perception of muscle soreness[1,2]. However all muscle actions (concentric, eccentric, static), appear to be capable of damaging muscle[1]. This damage can be specific to just a few macromolecules of tissue or result in large tears in the z-disk[3], sarcolemma[4,5], basal lamina[6], and supportive connective tissue[7], and induce injury to contractile elements and the cytoskeleton[8-13].

Three cytoplasmic isoforms of CK have been identified: CK-MM, CK-MB, and CK-BB. CK-MM is located in several domains of muscle fibers, principally in areas where ATP con-

sumption is high. More specifically, a substantial fraction (5-10%) of CK-MM is bound to the myofibrillar M-line structure by pairs of lysine residues[2]. In addition to the three cytoplasmic isoforms of CK, there are two mitochondrial isoenzymes (sarcomeric and non-sarcomeric).

Because of their distribution, the different CK isoforms provide specific information about the location of tissue injury. For example, circulating CK-MB rises after acute myocardial infarction[14]. Brain damage results in a rise CK-BB[15]. Circulating mitochondrial CK increases as a result of mitochondrial cytopathies[16]. The CK-MM isoform is a marker of myopathies[17] or exercise-induced muscle damage[2].

Vigorous exercise can result in increases in circulating CK. Heavy exercise, most notably eccentric muscle actions (lengthening contractions), often results in perforations in the sarcolemma and damage to sarcomeres[18]. Rises in circulating CK occur when the sarcolemma and Z-disks are damaged[2,19,20]. Exercise damages these structures when loading exceeds limits the muscle is accustomed to, and a resulting increase in membrane permeability is observed[18]. The increased membrane permeability allows CK to leak into interstitial fluid, where it then enters circulation via the lymphatic system[21]. Thus there is an intensity threshold which exercise must exceed for there to be substantial rises in CK[2]. Typically, serum CK activity rises within a few

Work	Classification (U.L⁻¹)					Decision Criteria for Classification	Exercise Protocol
	Low response	Medium response	Normal response	High response	Higher response		
Clarkson et al 1992	<500	500-2000	No	<2000	No	Discretionary	
Chen 2006	<500	500-2000	No	2000-10000	>10000	Discretionary	30 eccentric contractions with the elbow flexors of nondominant arm.
Heled et al 2007	No	No	<230	≥230	No	$\Delta CK \geq 90^{th}$ percentile	stepping up and down two stairs (30-cm height each) for 5 min at a pace of 54 steps/min followed by 15 knee bends completed within 1 min (3-s count down and 2-s count up were done to increase the eccentric contraction time). A backpack weighted at 30% of their body weight was worn during both tests.
Totsuka et al 2002	<300	No	No	>500	No	Discretionary	90 min of bicycling at a set absolute workload (1.5 kp, at 60 rpm) on 3 consecutive days
Machado & Willardson, 2010	No	No	<556.2[1] and <442.3[2]	≥556.2[1] and ≥442.3[2]	No	$\Delta CK \geq 90^{th}$ percentile	Three sets with 10RM loads were completed for the chest press, cable pulldown, biceps curl, triceps extension, leg extension, and prone leg curl
Do Carmo et al 2011	No	No	<475.1	≥475.1	No	CKpeak $\geq 90^{th}$ percentile	4 sets of biceps curl at 85% of 1-RM with 1 minute rest interval length between sets. The subjects were instructed to extend the elbows from an elbow flexed (50°) to an extended position (170°) and return to the flexed position in 3 s (~1 s to concentric and ~2 s to eccentric phase).
[1]CKpeak1; [2]CKpeak3, referring to resistance exercise with 1 min and 3 min rest intervals, respectively.							

Table 1. Sample classification schemes and criteria for categorizing creatine kinase response to resistance exercise.

hours after resistance exercise. From normal resting ranges of 60-400 U·L⁻¹ (see Schlattner et al.[22]), CK activity rises by ~100% within 8h after resistance exercise. CK activity continues to rises reaching levels ranging from 300-6000+ U·L⁻¹, with peak levels seen anywhere from 24 to 96 h after the initial exercise bout[2]. The location of that intensity threshold to induce a rise in CK, the time course of the rise in CK, and the extent to which CK rises after exercise, are highly variable and affected by individual factors and exercise variables[23-26].

Individual factors associated with elevated CK

High-responders and genotype

Some individuals studied have been classified as "high-responders" (HR) in light of the much higher rise in CK after resistance exercise as compared to an average, or normal response (NR). Unfortunately, there is no consensus regarding a clinical definition of CK activity to establish an individual as being HR. The distinction between HR and NR has thus far

been operationally defined by individual experiments, for example, Heled et al.[24] categorized high-responders as those who displayed a post-exercise change in CK≥ the 90th percentile of their cohort. Clarkson et al.[27] defined three groups, low (LR), medium (MR) and high (HR), based upon the amount of increase in CK. The LR where those having a peak CK of less than 500 U.l⁻¹, the MR between 500 and 2,000 U.l⁻¹, and the HR were those having a peak CK response of over 2,000 U.l⁻¹. More recently, Chen[28] created another group: higher responders (HrR) those exceeding 10,000 U.l⁻¹). It is important to note that this classification diversity could be associated with exercise protocol differences rather than individual (biological) differences. Heled et al.[24] used a low intensity, high volume exercise stimulus, while Cleak and Eston[29] and Chen[28] used a vigorous eccentric exercise protocol. It is possible that the exercise protocol used by Heled et al.[24] would not have induced muscle damage to the magnitude of those experiments employing eccentric actions, allowing the classification of subjects into only two groups (Table 1).

However it is defined, the difference between HR and NR

is thought to be, at least in part, due to genetic variation. Several investigations have sought to identify specific genetic markers for HR. So far, several genetic polymorphisms have been identified as being related to HR, including chemokine ligand 2 and chemokine receptor 2[30], myosin light chain kinase[31], insulin-like growth factor II[32], CK-MM[24] and the ACE genotypes[33].

α-actinin-3 is an actin-binding protein present in skeletal muscle and represents a major structural component of the Z line in the sarcomere[34]. Owing to its location, it was postulated that this protein may play a role in maintenance of the structural integrity of sarcomeres and muscle cells during eccentric muscle contractions[35].

Interestingly, the synthesis of α-actinin-3 is encoded for the ACTN3 gene (11q13–q14)[34], which has shown a single nucleotide polymorphism (SNP), called ACTN3 R577X polymorphism, with clinical relevance. Homozygosity of the X-allele results in an absence of ACTN3 expression, with no apparent association with muscle disease phenotypes[36]. Despite this, it has been postulated that the protein may play a role in maintenance of the structural integrity of sarcomeres and muscle cells during eccentric muscle contractions[34,35].

Vincent et al.[34] showed that subjects homozygous to the X-allele tended to have higher peak CK values after eccentric exercise when compared to those with a homozygous R-allele. The authors hypothesized that ACTN3-deficient fibers, as observed in homozygous X-allele individuals, could increase muscle damage, inducing a greater increases in CK response to eccentric exercise. Similar results were found by Pimenta et al.[37] when studying soccer professional athletes.

Body composition and sex

Other individual factors that may influence CK response include body composition and sex. In regard to body composition, a higher percent body fat has been found to be related to HR while body mass index was not[24]. It is possible that a higher body fat may provide an indicator of a phenotype related to an intricate relationship between genotypic profile and low habitual physical activity levels, giving poorly conditioned muscles, and then, lead to higher susceptibility to muscle damage after exercise.

Sex may also influence the extent of serum CK increase after exercise. In animal studies, females consistently appear to be less susceptible to muscle damage than males. It appears that estrogen has strong antioxidant properties that help maintain muscle cell membrane permeability after exercise[38], resulting in a lower rise in serum CK[39]. However, the available human studies have produced markedly different results, where women have either shown no difference in post-exercise serum CK, or displayed higher elevation in serum CK than men[40].

Exercise factors related to CK

Mechanisms of exercise-induced muscle damage

Experimental evidence implicates two main stressors as causes of exercise-induced muscle damage: mechanical stress and metabolic stress. Mechanical stress placed on muscle during exercise, largely induced by stretching of sarcomeres, causes disruptions in the contractile apparatus, muscle cytoskeleton and sarcolemma-associated proteins[41]. Metabolic stress is placed on muscle during exercise due to free radical formation and calcium overload. Elevation in O_2 consumption during exercise leading to increased activity in the electron transport chain, increased semiquinone in the mitochondria and xanthine oxidase in capillary endothelial cells[42], all of which may lead to increased production of free radicals and consequent damage to cell membranes. Further, muscle may experience an ischemia/reperfusion-like state in the transition from exercise to recovery[43], which would further enhance free radical production[44,45]. Calcium levels increase within resting muscle fibers after eccentric contractions, migrating from stretch-activated calcium channels[46], damaged transverse tubules[47], and possibly the sarcoplasmic reticulum. This calcium influx consequently activates proteases, phospholipases, lysosomal enzymes, and calpains, all of which increase protein turnover in muscle. Calpains, in particular, are thought to be a primary mediator of muscle damage after eccentric contractions[48]. Mechanical factors have been suggested as those most responsible for muscle injury[49].

Amount of work performed, surprisingly small effect

The amount of work performed during a resistance exercise bout is often expressed with the term *volume load*, defined as weight x repetitions x sets. Volume load yields the total tonnage lifted in an exercise session. It would seem logical to presume that a greater volume load lifted in a given exercise session would produce more trauma to the muscles, and thus a higher serum CK activity. Surprisingly, research to date indicates a more complicated relationship between work performed and the rise in serum CK.

Several investigations[25,50] have noted a positive relationship between volume load performed in a resistance exercise bout and serum CK. However, the correlations between the two are generally mild, for example, r=0.44 in one study[50].

A study by Nosaka and Clarkson[51] provides a classic illustration of the tenuous relationship between work and serum CK elevation. In this experiment, groups of subjects performed 24 maximal eccentric actions of the elbow flexors with either a single arm or both arms simultaneously on an isokinetic dynamometer. It was expected that the group performing exercise with two arms, exercising roughly twice the muscle mass as the comparison group, would display a greater rise in serum CK. Surprisingly, serum CK elevation was similar between groups[51]. Similarly, in diseases where serum CK elevation is associated with tissue damage, such as muscular dystrophy[52] and myocardial infarction[53], the extent of tissue damage is not strongly correlated with the rise of serum CK. Indeed, animal evidence suggests serum CK elevation is more strongly affected by lymph flow than the extent of tissue injury[54].

Shorter rest interval related to higher CK in some populations

The rest interval between sets of resistance exercise is an often overlooked factor in resistance exercise. However, sev-
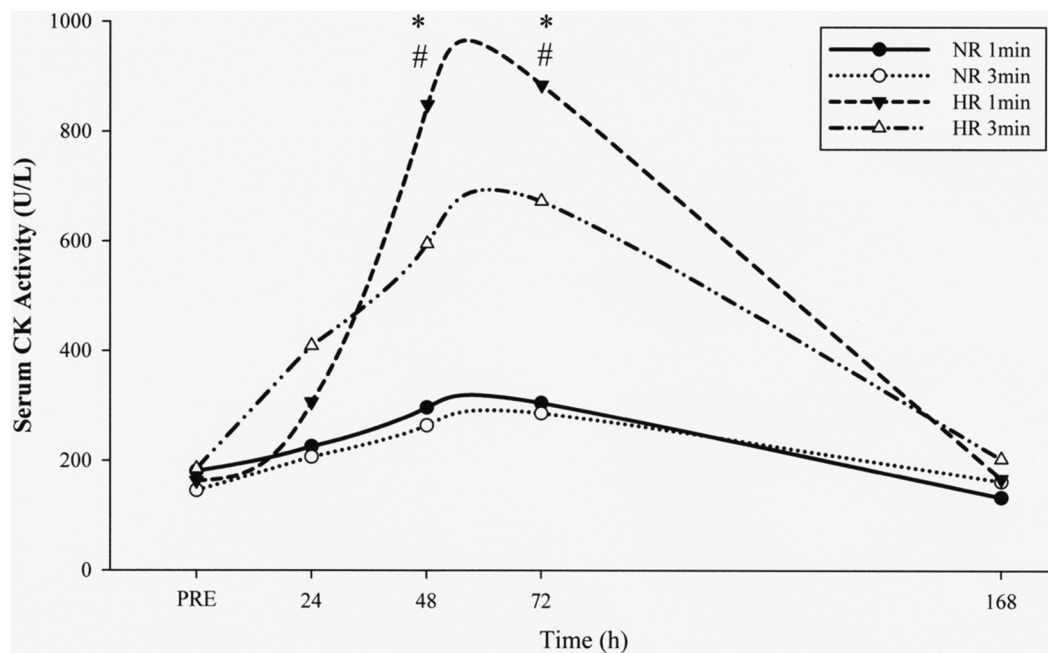
Figure 1. Serum CK activity before (PRE) and 24, 48, 72, and 168 H after resistance exercise with 1 min and 3 min rest inter-set rest intervals between NR (n=25) and HR (n=7). *HR group had significantly greater CK activity at the 1-versus 3-min bout ($P < 0.05$). #HR group had significantly greater CK activity versus the NR group at the 1- and 3-min bouts ($P<0.05$). Data originally published in Machado M & Willardson JM 2010. Short Recovery Augments Magnitude of Muscle Damage in High Responders. Med Sci Sports Exerc 42:1370-1374.

eral studies[55-57] have found that resistance exercise with short (60s) versus longer (180s) rest intervals between sets may produce a higher rise in serum CK.

Mayhew et al.[56] compared serum creatine kinase concentrations following two sessions that consisted of 10 sets of 10 repetitions at 65% 1RM in leg press with either 1 min or 3 min rest intervals between sets. Significant elevations in serum CK concentrations were demonstrated at 24 hours post-session for both rest conditions. However, serum CK was significantly greater when resting 1 min versus 3 min between sets. This occurred despite the fact that subjects lifted a significantly lower volume load during the 1 min rest protocol. In contrast when volume was held equal between rest conditions, studies have found no difference between shorter vs. longer rest intervals[25,58]. A possible explanation for these different findings might be that subjects in the latter two experiments were accustomed to training with 1-2 min rest intervals between sets before the experiment. However, subjects in Mayhew's[56] study were not.

Training evokes fairly rapid adaptations to exercise. For example, Buresh et al.[59] measured the hormonal response to resistance exercise between groups employing short (1 min) versus long (2.5 min) rest intervals longitudinally, over the course of a 10-week training cycle. While the short rest interval group displayed a significantly higher elevation in post-exercise testosterone and cortisol than was seen in the long rest interval group after one week of training, these differences had disappeared by five weeks of training. Thus it appears that subjects can quickly adapt to training at a specific rest interval, after which post-exercise increases in circulating hormones (and perhaps enzymes) will depend less on the rest interval employed and more on the total amount of work performed. Indeed, such an adaptation would also explain the similarity in CK activity despite differing rest intervals[25,58].

Machado and Willardson[55] compared 60 vs. 180s rest intervals in a trained population without equalizing volume between the conditions. Unsurprisingly, subjects lifted a lesser volume load under the conditions with shorter rest. Interestingly, they distinguished subjects' CK responses between HR and NR. It was found that short rest intervals led to a significantly higher CK activity in HR subjects, but were no different from long rest intervals in NR[55]. Thus there is evidence to support that a shorter rest interval may evoke a higher CK response to resistance exercise, at least in some (HR) individuals (Figure 1).

Movement speed

Relatively few studies have examined the effect of velocity of movement on rises in serum CK. One recent study found fast velocity ($210°·s^{-1}$) eccentric contractions produced a 4.5 higher serum CK activity than slower ($30°·s^{-1}$) actions given for the same time under tension[60]. Given the positive relationship between force and velocity for eccentric contractions, this finding is understandable. Indeed, fast ($180°·s^{-1}$) velocity ec-

centric contractions have also been shown to decrease post-exercise torque, activate protein degradation signaling pathways (FOXO1 and FOXO3) and elevate myostatin content[61] in rat skeletal muscle. These changes, characterized as atrophic[61], may contribute to higher CK activity as well. For concentric muscle actions, in which force and velocity are inversely related, it is likely that a slower movement speed would produce higher elevations in serum CK.

Muscle group - upper body

Several recent studies[62-65] have compared upper and lower body exercise, consistently finding of greater elevations in soreness and serum CK following upper body exercise compared to lower body exercise. Most notably, Chen et al.[62] recent experiment employed the same subjects exercising different (elbow vs. knee, extensor vs. flexor) muscle groups repeatedly. Interestingly, they observed that elevated CK responses occurred after exercise of some muscle groups, but not others, within the same subject. Thus, there is strong evidence to support that the muscle group exercised may be the most important factor in determining CK response.

Exactly why upper body exercise induces a greater rise in CK is still unknown. Jamurtas et al.[63] proposed that it is likely that people are more often exposed to eccentric actions using the muscles of their lower body in daily activities (i.e. descending stairs, sitting) than the muscles of their upper body.

Chen et al.[62] noted differences in markers in muscle damage not only between upper body and lower body muscles, but also in antagonist muscle groups around the same joint (e.g., knee extensor versus flexor). From these differences, they concluded that different characteristics of muscle damage between muscles are very specific, and suggested it is ideal to use specific exercise models of specific muscles to understand the nature of muscle damage for a given muscle group.

Strategies to reduce the rise in serum CK

Training status and repeated bout effect

The most prominent, controllable variable that alters the magnitude of serum CK rise after exercise is familiarization with the activity. Sedentary individuals display greater increases in CK following resistance exercise[66]. However, one single exposure to a bout of resistance exercise diminishes the rise in CK (and muscle soreness and other markers of muscle damage) to a subsequent bout of the same exercise. This phenomenon has been termed "the repeated bout effect", and has been demonstrated to occur in many studies[28,67,68]. The protective effect gained from one bout of exercise is developed within a few days following the initial damaging event.

Adaptations behind the repeated bout effect are thought to include a shift towards greater recruitment of slow-twitch motor units, the generation of new sarcomeres in series, thereby reducing the extent of microtrauma, and a downregulation of inflammation, that would limit the extent of post-exercise cell damage in the days following the exercise[68,69]. The protective effect of one bout of exercise has been documented

to persist for as long as 6 months following the initial exposure[70]. Thus regular training should provide a powerful resistance to large rises in serum CK activity.

Aging and muscle fiber type

Several studies have compared exercise-induced muscle damage responses, including CK activity, among different age groups. Consistently, post-exercise CK activity has been found to be higher in young adults than in either children[71-73], or elderly subjects[73,74] following exercise of the same relative intensity.

A possible explanation for why young adults experience a greater rise in CK activity (as well as other markers of muscle damage) is the difference in muscle fiber type between age groups. Fast-twitch (type II) muscle are more susceptible to exercise-induced muscle damage[75] and more prevalent in adults than in either children or the elderly[76-78]. Alternately, the muscles of adults may be more susceptible to damage as they must generate a greater force per fiber during eccentric contractions, owing to the greater body mass of adults vs. children[71], and presumably, the elderly.

Interestingly, Gorianovas et al.[73] recently documented that young adults displayed a greater attenuation of CK activity (and other markers of muscle damage) after a second exposure to eccentric exercise[73]. In this study, boys (mean age= 11.8 y), young men (mean age= 20.8 y) and elderly men (mean age= 63.2 y) performed two bouts of 100 drop jumps from a 0.5 m height, separated by two weeks. Consistent with previous studies, the young men displayed higher CK activity, perceived soreness, and decreases in voluntarily and electrically-invoked torque than either the boys or elderly men after the first bout. However, the young men also displayed greater decreases in CK activity 24h (young men= -685.2 IU/L, boys= -286.4 IU/L, elderly men= -70.7 IU/L) and 48h (young men= -881.2 IU/L, boys= -205.7 IU/L, elderly men= -206 IU/L) after their second exposure of exercise, relative to the first bout. Thus, while exercise-induced muscle damage and CK activity are greater in young adults, young adults' muscles may also adapt more readily to exercise.

Nutritional intervention

Several studies have investigated the use of dietary supplements as a means to offset exercise-induced muscle damage, as measured by CK activity. Branched chain amino acid (BCAA) supplementation appears to offset rises in serum CK after exercise. BCAA supplementation in subjects who were already ingesting recommended BCAA levels in their diets reduced the rise in CK (and LDH) after either vigorous endurance exercise[79,80] or resistance exercise81.

It is suggested that phosphocreatine, due to its amphipathic nature, can bind to the plasma membrane increasing its stability[82,83] and reducing the CK release to serum. However, despite many effects of creatine supplementation, the experimental results are conflicting on the relationship between this supplementation and the integrity of the muscle macrostructure[84,85].

Beta hydroxymethylbuterate (HMB) supplementation has been investigated as a means to stabilize muscle cell mem-

branes and reducing the extent of post-exercise damage. Available studies find mixed results, with some reporting an attenuated rise in CK after exercise with HMB supplementation[86], while others report no impact[87] of HMB supplementation. A recent meta-analysis of HMB's effect on muscle damage concluded that the supplement's impact on CK was "unclear"[88]. Additionally, the use of antioxidant supplements has been proposed in recent years[89]. Experimentation with dietary supplements that may attenuate the extent of muscle damage is ongoing, and represents a dynamic and growing avenue of research.

Application of cold , heat, massage, and compression

Immersion in either cold or warm water, ice and compression have been traditionally applied as modalities to assist the recovery from exercise. Briefly cold is theorized to reduce post-exercise inflammation and edema, while heat increases circulation to facilitate the removal of waste products. Available studies indicate that cold application – in the form of cold water immersion[90], contrast (cold-hot) immersion[91] or whole body cryotherapy[92] can attenuate post-exercise rises in serum CK. A recent study of cold application during (in between sets of) resistance exercise found that while cold application increased the workload completed, it did not alter the rise of serum CK[93].

Fewer data support the application of warmth after exercise to reduce serum CK. A recent study of post-exercise warm-water immersion found it to be successful in dampening the rise of serum CK[94], however this experiment limited the time of observation to a 6-h post-exercise window, likely too short to observe peak increases in serum CK. Pre-exercise warming has been proposed as a method to blunt exercise-induced muscle damage, possibly by decreasing muscle viscosity or increasing elasticity of the musculotendinous unit. Studies of pre-exercise immersion in warm water have found it may significantly blunt the rise of serum CK[95], though this finding is inconsistent[96].

Massage after exercise often reduces the post-exercise rise in serum CK, compared to that in a control[97,98], though not always[99]. Compression garments have also been found to effectively reduce the rise in serum CK after exercise[100,101]. Studies of ice massage have yielded mixed results[102,103]. In sum, cold therapy, massage, and compression have been found to attenuate post-exercise rises in serum CK. Less evidence supports the tactic of pre-exercise warming to attenuate rises in serum CK.

Concluding remarks

Creatine kinase is one of several (LDH, myoglobin, troponin, etc.) markers of exercise-induced muscle damage. In general, studies have found serum CK activity after exercise to poorly related to functional measures of muscle soreness, strength, range of motion, etc[104]. Post-exercise losses in strength are not coupled to the rise in CK[105].

Interpretation of the meaning of CK and other circulating muscle proteins is challenging. Often, a lower rise of CK (or Mb or LDH) after exercise is equated to a less traumatic exercise session, but not always. For example, Viitasalo et al.[106], found subjects treated with warm jet massage during vigorous power training retained continuous jumping power above levels seen in a control state. However, the authors attributed this observation in part to an increased Mb release seen during the massage treatment. They interpreted the increased release of proteins from muscle as being helpful in the maintenance of neuromuscular performance[106]. Given the poor relation to functional outcomes, and the question of how to interpret CK's rise in circulation after exercise, CK appears to be of more use as a qualitative marker that some trauma to skeletal muscle has occurred, rather than a quantitative indicator of the extent of muscle damage.

Based on the available studies, individual differences and exercise variables highly contribute to the extent of CK accumulation. For individual factors, several polymorphisms in genotype that affect the rise in CK have been identified and research to identify more is ongoing. In regards to exercise programming, it appears that a high volume of upper-body exercise, with short rest intervals taken between sets, would tend to produce the greatest increase in CK. Whether this type of protocol would affect clinical outcomes, such as an increased risk exertional rhabdomyolysis is questionable, as there is currently no established link between an exaggerated CK response and exertional rhabdomyolysis. Interestingly, a recent case in the United States, in which 14 high school American football players experienced rhabdomyolysis consisted of just such an exercise bout. During summer training these athletes performed an exercise bout consisting of two upper body exercises: pushups and chair dips. The exercises were performed with virtually no rest, as supersets, and continuously for work intervals of 30s each, and then repeated with decreasing work times until a final work set of 5s each was reached. The consequences of this exercise bout resulted in hospitalization of 14 players, three of whom required an emergency fasciotomy of their Triceps Brachii[107].

Noakes[108] proposed that subjects who displayed abnormally large increases in CK after exercise may have some unrecognized subclinical myopathy. Subsequent work has supported his assertion, finding exaggerated CK responses in conditions such as McCardle's disease[109] and calveolinopathies[110,111]. Thus evaluation of CK pre- and post-exercise may provide a diagnostic tool for the detection of myopathies, with much less invasiveness than required of a muscle biopsy. This may be the most meaningful use of serum CK monitoring.

References

1. Clarkson PM, Byrnes WC, McCormick KM, Turcotte LP, White JS. Muscle soreness and serum creatine kinase activity following isometric, eccentric, and concentric exercise. Int J Sports Med 1986;7:152-155.
2. Brancaccio P, Maffulli N, Buonauro R, Limongelli FM. Serum enzyme monitoring in sports medicine. Clin Sports

Med 2008;27:1-18.

3. Lieber RL, Friden J. Selective damage of fast glycolytic muscle fibres with eccentric contraction of the rabbit tibialis anterior. Acta Physiol Scand 1988;133:587-8.

4. Duan C, Delp MD, Hayes DA, Delp PD, Armstrong RB. Rat skeletal muscle mitochondrial [Ca2+] and injury from downhill walking. J Appl Physiol 1990;68:1241-51.

5. Takekura H, Fujinami N, Nishizawa T, Ogasawara H, Kasuga N. Eccentric exercise-induced morphological changes in the membrane systems involved in excitation-contraction coupling in rat skeletal muscle. J Physiol 2001; 533:571-583.

6. Koskinen SO, Wang W, Ahtikoski AM, Kjaer M, Han XY, Komulainen J, Kovanen V, Takala TE. Acute exercise induced changes in rat skeletal muscle mRNAs and proteins regulating type IV collagen content. Am J Physiol Regul Integr Comp Physiol 2001;280:R1292-300.

7. Stauber WT, Clarkson PM, Fritz VK, Evans WJ. Extracellular matrix disruption and pain after eccentric muscle action. J Appl Physiol 1990;69:868-874.

8. Fridén J, Kjörell U, Thornell LE. Delayed Muscle Soreness and Cytoskeletal Alterations: An Immunocytological Study in Man. Int J Sports Med 1984;05:15-18.

9. Fridén J, Seger J, Ekblom B. Sublethal muscle fibre injuries after high-tension anaerobic exercise. Eur J Appl Physiol 1988;57:360-368.

10. Friden J, Lieber RL. Ultrastructural evidence for loss of calcium homeostasis in exercised skeletal muscle. Acta Physiol Scand 1996;158,381-2.

11. Lieber RL, Thornell LE, Fridén J. Muscle cytoskeletal disruption occurs within the first 15 min of cyclic eccentric contraction. J Appl Physiol 1996;80:278-84.

12. Roth SM, Martel GF, Ivey FM, Lemmer JT, Metter EJ, Hurley BF, Rogers MA. High-volume, heavy-resistance strength training and muscle damage in young and older women. J Appl Physiol 2000;88:1112-1118.

13. Roth SM, Martel GF, Ivey FM, Lemmer JT, Tracy BL, Hurlbut DE, Metter EJ, Hurley BF, Rogers MA. Ultrastructural muscle damage in young vs. older men after high-volume, heavy-resistance strength training. J Appl Physiol 1999;86:1833-1840.

14. Borrayo-Sanchez G, Sosa-Jarero F, Borja-Teran B, Isordia-Salas I, Arguero-Sanchez R. Qualitative determination of markers for myocardiac necrosis during pre-hospital admission for acute coronary syndrome. Cir Cir 2006;74:231-235.

15. Pfeiffer FE, Homburger HA, Yanagihara T. Creatine kinase BB isoenzyme in CSF in neurologic diseases. Measurement by radioimmunoassay. Arch Neurol 1983; 40:169-172.

16. Stadhouders AM, Jap PH, Winkler HP, Eppenberger HM, Wallimann T. Mitochondrial creatine kinase: a major constituent of pathological inclusions seen in mitochondrial myopathies. PNAS 1994;91:5089-5093.

17. Nigro G, Comi LI, Limongelli FM, Giugliano MA, Politano L, Petretta V, Passamano L, Stefanelli S. Prospective study of X-linked progressive muscular dystrophy in Campania. Muscle Nerve 1983;6:253-262.

18. Clarkson PM, Sayers SP. Etiology of exercise-induced muscle damage. Can J Appl Physiol 1999;24:234-48.

19. Brancaccio P, Maffulli N, Limongelli FM. Creatine kinase monitoring in sport medicine. Br Med Bull 2007;81-82:209-230.

20. Brancaccio P, Lippi G, Maffulli N. Biochemical markers of muscular damage. Clin Chem Lab Med 2010;48:757-67.

21. Bijsterbosch MK, Duursma AM, Smit MJ, Bos OJ, Bouma JM, Gruber M. Several dehydrogenases and kinases compete for endocytosis from plasma by rat tissues. Biochem J 1985;229:409-417.

22. Schlattner U, Tokarska-Schlattner M, Wallimann T. Mitochondrial creatine kinase in human health and disease. Biochim Biophys Acta 2006;1762:164-180.

23. Carmo FC, Pereira R, Machado M. Variability in resistance exercise induced hyperCKemia. Isok Exerc Sci 2011;19:191-197.

24. Heled Y, Bloom MS, Wu TJ, Stephens Q, Deuster PA. CK-MM and ACE genotypes and physiological prediction of the creatine kinase response to exercise. J Appl Physiol 2007;103:504-510.

25. Machado M, Koch AJ, Willardson JM, Pereira LS, Cardoso MI, Motta MK, Pereira R, Monteiro AN. Effect of varying rest intervals between sets of assistance exercises on creatine kinase and lactate dehydrogenase responses. J Strength Cond Res 2011;25:1339-1345.

26. Machado M, Pereira R, Willardson JM. Short intervals between sets and individuality of muscle damage response. J Strength Cond Res 2012;26:2946-52.

27. Clarkson PM. Exercise-induced muscle damage-animal and human models. Med Sci Sports Exerc 1992;24:510-511.

28. Chen TC. Variability in muscle damage after eccentric exercise and the repeated bout effect. Res Q Exerc Sport 2006;77:362-371.

29. Cleak MJ, Eston RG. Muscle soreness, swelling, stiffness and strength loss after intense eccentric exercise. Br J Sports Med 1992;26:267-272.

30. Hubal MJ, Devaney JM, Hoffman EP, Zambraski EJ, Gordish-Dressman H, Kearns AK, Larkin JS, Adham K, Patel RR, Clarkson PM. CCL2 and CCR2 polymorphisms are associated with markers of exercise-induced skeletal muscle damage. J Appl Physiol 2010;108:1651-1658.

31. Clarkson PM, Hubal MJ. Exercise-induced muscle damage in humans. Am J Phys Med Rehabil 2002;81:S52-69.

32. Devaney JM, Hoffman EP, Gordish-Dressman H, Kearns A, Zambraski E, Clarkson PM. IGF-II gene region polymorphisms related to exertional muscle damage. J Appl Physiol 2007;102:1815-1823.

33. Yamin C, Amir O, Sagiv M, Attias E, Meckel Y, Eynon N, Sagiv M, Amir RE. ACE ID genotype affects blood creatine kinase response to eccentric exercise. J Appl Physiol 2007;103:2057-2061.

34. Vincent B, Windelinckx A, Nielens H, Ramaekers M, Van

Leemputte M, Hespel P, Thomis MA. Protective role of α-actinin-3 in the response to an acute eccentric exercise bout. J Appl Physiol 2010;109:564-573.

35. MacArthur DG, North KN. A gene for speed? The evolution and function of alpha-actinin-3. Bioessays 2004; 26:786-795.

36. Vainzof M, Costa CS, Marie SK, Moreira ES, Reed U, Passos-Bueno MR, Beggs AH, Zatz M. Deficiency of alpha-actinin-3 (ACTN3) occurs in different forms of muscular dystrophy. Neuropediatrics 1997;28:223-228.

37. Pimenta EM, Coelho DB, Cruz IR, Morandi RF, Veneroso CE, Pussieldi GA, Carvalho MRS, Silami-Garcia E, Fernández JAP. The ACTN3 genotype in soccer players in response to acute eccentric training. Eur J Appl Physiol 2012;112:1495-1503.

38. Tiidus PM. Influence of estrogen on muscle plasticity. Braz J Biomotricity 2011;4:143-155.

39. Tiidus PM. Estrogen and gender effects on muscle damage, inflammation, and oxidative stress. Can J Appl Physiol 2000;25:274-287.

40. Clarkson PM, Hubal MJ. Are women less susceptible to exercise-induced muscle damage? Curr Opin Clin Nutr Metab Care 2001;4:527-531.

41. Friden J, Lieber RL. Eccentric exercise-induced injuries to contractile and cytoskeletal muscle fibre components. Acta Physiol Scand 2001;171:321-326.

42. Su QS, Zhang JG, Dong R, Hua B, Sun JZ. Comparison of changes in markers of muscle damage induced by eccentric exercise and ischemia/reperfusion. Scand J Med Sci Sports 2010;20:748-756.

43. Tamaki T, Uchiyama S, Tamura T, Nakano S. Changes in muscle oxygenation during weight-lifting exercise. Eur J Appl Physiol Occup Physiol 1994;68:465-469.

44. Garlick PB, Davies MJ, Hearse DJ, Slater TF. Direct detection of free radicals in the reperfused rat heart using electron spin resonance spectroscopy. Circ Res 1987;61:757-760.

45. Jassem W, Fuggle SV, Rela M, Koo DD, Heaton ND. The role of mitochondria in ischemia/reperfusion injury. Transplantation 2002;73:493-499.

46. Sonobe T, Inagaki T, Poole DC, Kano Y. Intracellular calcium accumulation following eccentric contractions in rat skeletal muscle in vivo: role of stretch-activated channels. Am J Physiol Regul Integr Comp Physiol 2008; 294:R1329-1337.

47. Takekura H, Fujinami N, Nishizawa T, Ogasawara H, Kasuga N. Eccentric exercise-induced morphological changes in the membrane systems involved in excitation-contraction coupling in rat skeletal muscle. J Physiol 2001;533:571-583.

48. Zhang BT, Yeung SS, Allen DG, Qin L, Yeung EW. Role of the calcium-calpain pathway in cytoskeletal damage after eccentric contractions. J Appl Physiol 2008; 105:352-357.

49. Lieber RL, Friden J. Mechanisms of muscle injury gleaned from animal models. Am J Phys Med Rehabil 2002;81:S70-79.

50. Machado M, Willardson JM, Silva DP, Frigulha IC, Koch AJ, Souza SC . Creatine Kinase Activity Weakly Correlates to Volume Completed Following Upper Body Resistance Exercise. Res Q Exerc Sport 2012;83:276-281.

51. Nosaka K, Clarkson PM. Relationship between post-exercise plasma CK elevation and muscle mass involved in the exercise. Int J Sports Med 1992;13:471-475.

52. Kroll M, Otis J, Kagen L. Serum enzyme, myoglobin and muscle strength relationships in polymyositis and dermatomyositis. J Rheumatol 1986;13:349-355.

53. Roe CR, Cobb FR, Starmer CF. The relationship between enzymatic and histologic estimates of the extent of myocardial infarction in conscious dogs with permanent coronary occlusion. Circulation 1977;55:438-449.

54. Komulainen J, Takala TE, Vihko V. Does increased serum creatine kinase activity reflect exercise-induced muscle damage in rats?" Int J Sports Med 1995;16:150-154.

55. Machado M, Willardson JM. Short recovery augments magnitude of muscle damage in high responders. Med Sci Sports Exerc 2010;42:1370-1374.

56. Mayhew DL, Thyfault JP, Koch AJ. Rest-interval length affects leukocyte levels during heavy resistance exercise. J Strength Cond Res 2005;19:16-22.

57. Rodrigues BM, Dantas E, de Salles BF, Miranda H, Koch AJ, Willardson JM, and Simao R. Creatine kinase and lactate dehydrogenase responses after upper-body resistance exercise with different rest intervals. J Strength Cond Res 2010;24:1657-62.

58. Ribeiro V, Pereira R, Machado M. Resistance exercise-induced microinjuries do not depend on 1 or 3 minutes rest time interval between series. Int J Sports Sci 2008; 13:44-53.

59. Buresh R, Berg K, French J. The effect of resistive exercise rest interval on hormonal response, strength, and hypertrophy with training. J Strength Cond Res 2009;23:62-71.

60. Chapman D, Newton M, Sacco P, Nosaka K. Greater muscle damage induced by fast versus slow velocity eccentric exercise. Int J Sports Med 2006;27:591-598.

61. Ochi E, Hirose T, Hiranuma K, Min SK, Ishii N, Nakazato K. Elevation of myostatin and FOXOs in prolonged muscular impairment induced by eccentric contractions in rat medial gastrocnemius muscle. J Appl Physiol 2010;108:306-313.

62. Chen TC, Lin KY, Chen HL, Lin MJ, Nosaka K. Comparison in eccentric exercise-induced muscle damage among four limb muscles. Eur J Appl Physiol 2011; 111:211-223.

63. Jamurtas AZ, Theocharis V, Tofas T, Tsiokanos A, Yfanti C, Paschalis V, Koutedakis Y, Nosaka K. Comparison between leg and arm eccentric exercises of the same relative intensity on indices of muscle damage. Eur J Appl Physiol 2005;95:179-185.

64. Saka T, Bedrettin A, Yazici Z, Sekir U, Gur H, Ozarda Y. Differences in the magnitude of muscle damage between elbow flexors and knee extensors eccentric exercises. J

Sports Sci Med 2009;8:107-115.

65. Machado M, Brown LE, Augusto-Silva P, Pereira R. Is exercise-induced muscle damage susceptibility body segment dependent? Evidence for whole body susceptibility. J Musculoskelet Neuronal Interact 2013;13:105-110.

66. Vincent HK, Vincent KR. The effect of training status on the serum creatine kinase response, soreness and muscle function following resistance exercise. Int J Sports Med 1997;18:431-437.

67. Nosaka K, Aoki MS. Repeated bout effect: research update and future perspective. Braz J Biomotricity 2011;5:5-15.

68. McHugh MP. Recent advances in the understanding of the repeated bout effect: the protective effect against muscle damage from a single bout of eccentric exercise. Scand J Med Sci Sports 2003;13:88-97.

69. Stupka N, Tarnopolsky MA, Yardley NJ, Phillips SM. Cellular adaptation to repeated eccentric exercise-induced muscle damage. J Appl Physiol 2001;91:1669-1678.

70. Nosaka K, Clarkson PM, McGuiggin ME, Byrne JM. Time course of muscle adaptation after high force eccentric exercise. Eur J Appl Physiol Occup Physiol 1991; 63:70-76.

71. Webber LM, Byrnes WC, Rowland TW, Foster VL. Serum creatine kinase activity and delayed onset muscle soreness in children: a preliminary study. Pediatric Exercise Science 1989;1:351-359.

72. Soares JMC, Mota P, Duarte JA, Appell HJ. Children are less susceptible to exercise-induced muscle damage than adults: a preliminary investigation. Pediatric Exercise Science 1996;8:361-367.

73. Gorianovas G, Skurvydas A, Streckis V, Brazaitis M, Kamandulis S, McHugh MP. Repeated bout effect was more expressed in young adult males than in elderly males and boys. Biomed Res Int 2013;218970.

74. Lavender AP, Nosaka K. Comparison between old and young men for changes in makers of muscle damage following voluntary eccentric exercise of the elbow flexors. Appl Physiol Nutr Metab 2006;31:218-225.

75. Friden J, Seger J, Ekblom B. Sublethal muscle fibre injuries after high-tension anaerobic exercise. Eur J Appl Physiol Occup Physiol 1988;57:360-368.

76. Lexell J, Sjostrom M, Nordlund AS, Taylor CC. Growth and development of human muscle: a quantitative morphological study of whole vastus lateralis from childhood to adult age. Muscle a d Nerve 1992;15:404-409.

77. Kriketos AD, Baur LA, O'Connor J, Carey D, King S, Caterson ID, Storlien LH. Muscle fibre type composition in infant and adult populations and relationships with obesity. Int J Obes Relat Metab Disord 1997;21:796-801.

78. Brunner F, Schmid A, Sheikhzadeh A, Nordin M, Yoon J, Frankel V. Effects of aging on Type II muscle fibers: a systematic review of the literature. J Aging Phys Act 2007;15:336-348.

79. Coombes JS, McNaughton LR. Effects of branched-chain amino acid supplementation on serum creatine kinase and lactate dehydrogenase after prolonged exercise. J Sports Med Phys Fitness 2000;40:240-246.

80. Matsumoto K, Koba T, Hamada K, Sakurai M, Higuchi T, Miyata H. Branched-chain amino acid supplementation attenuates muscle soreness, muscle damage and inflammation during an intensive training program. J Sports Med Phys Fitness 2009;49:424-431.

81. Sharp CP, Pearson DR. Amino acid supplements and recovery from high-intensity resistance training. J Strength Cond Res 2010;24:1125-1130.

82. Matthews RT, Yang L, Jenkins BG, Ferrante RJ, Rosen BR, Kaddurah-Daouk R, Beal MF. Neuroprotective effects of creatine and cyclocreatine in animal models of Huntington's disease. J Neurosci 1998;18:156-163.

83. Saks VA, Strumia E. Phosphocreatine: molecular and cellular aspects of the mechanism of cardioprotective action. Curr Ther Res 1993;53:565-598.

84. Machado M, Pereira R, Sampaio-Jorge F, Knifis F, Hackney AC. Creatine supplementation: Effects on blood Creatine Kinase activity responses to resistance exercise and creatine kinase activity measurement. Brazilian J Pharm Sci 2009;45:751-757.

85. Rawson ES, Conti MP, Miles MP. Creatine supplementation does not reduce muscle damage or enhance recovery from resistance exercise. J Strength Cond Res 2007; 21:1208-1213.

86. van Someren KA, Edwards AJ, Howatson G. Supplementation with beta-hydroxy-beta-methylbutyrate (HMB) and alpha-ketoisocaproic acid (KIC) reduces signs and symptoms of exercise-induced muscle damage in man. Int J Sport Nutr Exerc Metab 2005;15:413-424.

87. Hoffman JR, Cooper J, Wendell M, Im J, Kang J. Effects of beta-hydroxy beta-methylbutyrate on power performance and indices of muscle damage and stress during high-intensity training. J Strength Cond Res 2004;18:747-752.

88. Rowlands DS, Thomson JS. Effects of beta-hydroxy-beta-methylbutyrate supplementation during resistance training on strength, body composition, and muscle damage in trained and untrained young men: a meta-analysis. J Strength Cond Res 2009;23:836-846.

89. Bowtell JL, Sumners DP, Dyer A, Fox P, Mileva KN. Montmorency cherry juice reduces muscle damage caused by intensive strength exercise. Med Sci Sports Exerc 2011;43:1544-1551.

90. Ascensao A, Leite M, Rebelo AN, Magalhaes S, Magalhaes J. Effects of cold water immersion on the recovery of physical performance and muscle damage following a one-off soccer match. J Sports Sci 2011;29:217-225.

91. Pournot H, Bieuzen F, Duffield R, Lepretre PM, Cozzolino C, Hausswirth C. Short term effects of various water immersions on recovery from exhaustive intermittent exercise. Eur J Appl Physiol 2011;111:1287-1295.

92. Banfi G, Lombardi G, Colombini A, Melegati G. Whole-body cryotherapy in athletes. Sports Med 2010;40:509-517.

93. Galoza P, Sampaio-Jorge F, Machado M, Fonseca R, and Sillva PAV. Resistance Exercise Inter-Set Cooling Strat-

egy: Effect on Performance and Muscle Damage. Int J Sports Physiol and Perf 2011;6:580-4.

94. Hassan ES. Thermal therapy and delayed onset muscle soreness. J Sports Med Phys Fitness 2011;51:249-254.

95. Skurvydas A, Kamandulis S, Stanislovaitis A, Streckis V, Mamkus G, Drazdauskas A. Leg immersion in warm water, stretch-shortening exercise, and exercise-induced muscle damage. J Athl Train 2008;43:592-599.

96. Nosaka K, Sakamoto K, Newton M, Sacco P. Influence of Pre-Exercise Muscle Temperature on Responses to Eccentric Exercise. J Athl Train 2004;39:132-137.

97. Smith LL, Keating MN, Holbert D, Spratt DJ, McCammon MR, Smith SS, Israel RG. The effects of athletic massage on delayed onset muscle soreness, creatine kinase, and neutrophil count: a preliminary report. J Orthop Sports Phys Ther 1994;19:93-99.

98. Zainuddin Z, Newton M, Sacco P, Nosaka K. Effects of massage on delayed-onset muscle soreness, swelling, and recovery of muscle function. J Athl Train 2005;40:174-180.

99. Schillinger A, Koenig D, Haefele C, Vogt S, Heinrich L, Aust A, Birnesser H, Schmid A. Effect of manual lymph drainage on the course of serum levels of muscle enzymes after treadmill exercise. Am J Phys Med Rehabil 2006;85:516-520.

100. Davies V, Thompson KG, Cooper SM. The effects of compression garments on recovery. J Strength Cond Res 2009;23:1786-1794.

101. Kraemer WJ, Flanagan SD, Comstock BA, Fragala MS, Earp JE, Dunn-Lewis C, Ho JY, Thomas GA, Solomon-Hill G, Penwell ZR, Powell MD, Wolf MR, Volek JS, Denegar CR, Maresh CM. Effects of a whole body compression garment on markers of recovery after a heavy resistance workout in men and women. J Strength Cond Res 2010;24:804-814.

102. Howatson G, Van Someren KA. Ice massage. Effects on exercise-induced muscle damage. J Sports Med Phys Fitness 2003;43:500-505.

103. Howatson G, Gaze D, van Someren KA. The efficacy of ice massage in the treatment of exercise-induced muscle damage. Scand J Med Sci Sports 2005;15:416-422.

104. Margaritis I, Tessier F, Verdera F, Bermon S, Marconnet P. Muscle enzyme release does not predict muscle function impairment after triathlon. J Sports Med Phys Fitness 1999;39:133-139.

105. Sewright KA, Hubal MJ, Kearns A, Holbrook MT, Clarkson PM. Sex differences in response to maximal eccentric exercise. Med Sci Sports Exerc 2008;40:242-251.

106. Viitasalo JT, Niemela K, Kaappola R, Korjus T, Levola M, Mononen HV, Rusko HK, Takala TE. Warm underwater water-jet massage improves recovery from intense physical exercise. Eur J Appl Physiol Occup Physiol 1995;71:431-438.

107. Russell M. Combination of intense drill, heat, dehydration, may have sent McMinnville players to hospital. In: The Oregonian; 2010.

108. Noakes TD. Effect of exercise on serum enzyme activities in human. Sports Med 1987;4:245-267.

109. Bruno C, Bertini E, Santorelli FM, DiMauro S. Hyper-CKemia as the only sign of McArdle's disease in a child. J Child Neurol 2000;15:137-138.

110. Carbone I, Bruno C, Sotgia F, Bado M, Broda P, Masetti E, Panella A, Zara F, Bricarelli FD, Cordone G, Lisanti MP, Minetti C. Mutation in the CAV3 gene causes partial caveolin-3 deficiency and hyperCKemia. Neurology 2000;54:1373-1376.

111. Woodman SE, Sotgia F, Galbiati F, Minetti C, Lisanti MP. Caveolinopathies: mutations in caveolin-3 cause four distinct autosomal dominant muscle diseases. Neurology 2004;62:538-543.