The Significance of Elevated Serum Enzyme Creatine Kinase as a Biomarker in Musculoskeletal Injury: A Review

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Abstract: Musculoskeletal injury (MSI) has been found to be associated with the abnormality in the serum enzyme creatine kinase (CK). When skeletal muscle sustains injury from the intense prolonged muscular contractions as a consequence of both pathological and physiological factors, it will cause an imbalance in cellular homeostasis increasing the membrane permeability to leak CK enzyme from cells into blood serum. This subsequent release of serum enzyme profusely into the plasma and not being falsely elevated by haemolysis has made CK the preferable biomarker than other serum enzymes. It is a key enzyme that catalyses the reversible transfer of a phosphoryl group (P) from phosphocreatine, producing ATP and creatine. It forms the core of an energy network found in both mitochondria and cytosol where the energy demands are high. Specifically, CK-MM is the type among major isozymes that is most widely distributed among highly differentiated skeletal muscle tissue. Injuries occur due to trauma, infections or even strenuous exercise to the skeletal muscles often trigger high levels of CK-MM. Thus, it is distinctly regarded as a fundamental biomarker for muscle injury. However, some studies show conflicts for lacking sensitivity and not establishing the proper extent of the injury. Accordingly, based on relevant current evidence and opinions, this review paper attempts to provide comprehensive importance of elevated serum enzyme CK as a biomarker for musculoskeletal injury (MSI) and examine if the elevated concentrations are useful for early detection of MSI.

Key Words: biomarker, creatine kinase, serum enzyme, musculoskeletal injury.

1. Introduction:

Musculoskeletal injury (MSI), simply refers to damage of muscular or skeletal systems, following intense activity as a consequence of both pathological and physiological factors [1]. It includes a wide range of inflammatory and degenerative conditions affecting the muscles, tendons, ligaments, joints, peripheral nerves, and supporting blood vessels [2]. Lately, it has become one of the leading causes of pain, suffering, and disability, deteriorating public health globally [3]. The use of biomarkers pertaining to early detection of MSI would be more reliable and efficient as the diagnostic approach.

Serum enzymes are extensively used as biomarkers for measuring the functional status of suspected musculoskeletal tissue injury [4,5]. Creatine kinase (CK), lactate dehydrogenase (LD), aspartate aminotransferase (AST), alanine aminotransferase (ALT), aldolase and pyruvate kinase are the abundantly found serum enzymes in the skeletal muscle, and these enzymes are released in substantial amount when an injury occurs in the skeletal muscle cell [6]. However, CK is the most preferred biomarkers used in the diagnosis of such injury. Unlike other serum enzymes, CK is readily released predominantly during cellular injury showing the greatest frequency of abnormality as well as is not falsely elevated by haemolysis that makes it most sensitive indicator among others [4,7,8].

The serum level of CK in the skeletal muscle cells varies widely in both pathological and physiological conditions. The cellular necrosis or the tissue damage either from acute or chronic injury can elevate the serum level of CK. This may be a result of both metabolic and mechanical
circumstances [6,7]. However, studies showing the fact that total CK can be affected by age, gender, race, muscle mass, physical activity and climatic condition also, can’t be ignored [10]. It is the main cause that affects the reference interval, as a result, normal range is very broadly skewed to the top end [10–14]. Thus, this review paper attempts to examine current evidence and opinions to elucidate the significance of elevated serum enzyme creatine kinase in relation to musculoskeletal injury (MSI) and examine if elevated concentrations are helpful for early detection.

2. Metabolism and function of Creatine kinase

a) Phosphocreatine + MgADP − − − H+ ⇌ MgATP2− + Creatine

b) Figure (1) a. Enzymatic reaction forming creatine kinase, b. In structural form [17].

Here, in this circuit of enzymatic reaction (Figure 1), the enzyme CK consumes ADP to catalyse the reversible transfer of a phosphoryl group (P) from phosphocreatinine, producing ATP and creatine [17–20]. Thus, Creatine kinase (also known as CK; adenosine-5’-triphosphate: creatine phosphotransferase; creatine phosphokinase; phosphocreatinine phosphokinase; creatine N-phosphotransferase; EC 2.7.3.2) is the key enzyme that forms the core of an energy network found in both mitochondria and cytosol where the energy demands are high like in muscle and brain [15,17,21].

CK is a dimeric globular protein comprising of two subunits with a molecular mass 43–45 kDa for each subunit [5,22]. A compact enzyme CK comprises of four major isozymes. Two are cytosolic and two are mitochondrial. In the cytosol, CK is composed of two tissue-specific subunit as M (muscle type) and B (brain type). These subunits allow the formation of three tissue-specific isozymes: CK-MB (cardiac muscle) existing as hetero-dimers, while CK-MM (skeletal muscle), and CK-BB (brain) prevailing as homo-dimers under physiological conditions [15,17,23]. The heterodimer form frequently appeared in a transitory fashion during the fetal and neonatal development of skeletal muscle, but also persisted, e.g. in rat and the human heart, throughout adult life [24]. The ubiquitous (Miu-CK) and the sarcomeric (Mis-CK) are the two mitochondrial isozymes usually existing as octamers but can be dissociated into dimmers [25]. The ubiquitous non-sarcomeric brain-type CK-BB is found in brain, heart, smooth muscle, nervous system, and other tissues, whereas the muscle-type CK (CK-MM) isoform is widely distributed among highly differentiated skeletal muscle tissue [26]. However, CK-MM is the specific key CK isozyme as it is the only isoform that interacts with the M-band region of a myofibrillar sarcomere [27].

All CK isoenzymes catalyze the reversible transfer of the γ-phosphate group of ATP to the guanidino group of Cr to yield ADP and PCr [28,29] as shown below in (Figure 2) [28]. This process enables muscle cells to build up a store of inorganic phosphate in the form of PCr which can be utilized for immediate ATP generation in periods of acute necessity. While this process can be sustained for up to 10 seconds in healthy
individuals, it provides gap until the body can begin to generate ATP through glycolytic and oxidative pathways [30,31]. The CK reaction also modulates intracellular pH to protect cells from damage associated with internal acidification and ATP depletion [32,33]. Thus, the CK acts not only acts as an energy buffer but also as a metabolic regulator [33]. Furthermore, PCr can interact with and protect cellular membranes [34], while Cr has been shown to scavenge free radicals and to harbor antioxidant properties [29,35,36].

![Figure 2. The creatine kinase (CK) reaction. PCr, phosphorylcreatine; Cr, creatine [28].](image)

Until the mid-1990s, serum enzyme CK levels were used as a key tool for diagnosing patients with myocardial infarction (MI). Consequently, the muscle protein troponin replaced the diagnostic role to a certain extent [17]. In recent years, biomarker serum enzyme CK has been given the special attention as the identification measure for the study of MIS i.e. muscle, tendon, and ligament [37,38]. In cell damage, muscle cell disruption, or disease, increased levels of serum CK have been identified. These cellular disturbances increase the membrane permeability which leads CK to leak from cells into blood serum [39]. Furthermore, many extensive research studies have shown the significance of serum enzyme CK profiles, still as an important indicator of the occurrence of musculoskeletal cell necrosis and tissue damage due to disease or trauma [10,17].

3. **Mechanism of musculoskeletal injury**

Muscles are prone to damage, like other body tissues. And skeletal muscle injury is the most common injuries that often challenge the health of human beings. It ranges from mild to severe that leads to minor significant pain and even to disability [40]. Crush injuries, contusion, laceration, electrical injury, freezing, or direct hits are the types of traumatic muscle injuries that can have long-term effects on the capacity of muscle to function well. While contraction-induced muscle injuries often result from demanding muscular work or strenuous exercise [40,41].

Normally, the biomechanics of MSI are dependent on the force that must be applied to do tasks, the duration of the force applied, and the frequency with which tasks are performed [42]. The inequality between the external load due to physical exertion and posture and the capability of the human body to endure that load often results in the MSI [43]. Though activities involving heavy force, repetition, or maintaining a strenuous static posture are the common risk factors for MSI, minimal force involving tasks or simple motion could trigger the onset of such injury [44], which could be the indication for pathological underlying causes. Furthermore, sports and work related activities are other contributing factors that account immense majority in MSIs [1,2], as these activities normally require severe exertion. The severe exertion in the body causes mechanical and thermal disturbances in the muscle with ATP depletion that triggers injury [45].

Skeletal muscle generally contracts and relaxes accordingly while functioning. Sarcomeres, the microscopic units found in the myofibrils of the muscle are responsible for muscle contraction and relaxation with the help of muscle protein called myosin that acts when stimulated by nerves. These sarcomeres also release (via the ryanodine receptor) and re-uptake (via the Ca2+-ATPase pump) Ca2+ during contraction and relaxation, respectively. But when the muscle is exposed to the immense force it cannot withstand, the tissue initiates to tear resulting in the muscle damage [46,47].

4. **Creatine Kinase in skeletal muscle**

In normal serum, total CK is available almost only in the MM fraction of skeletal muscle [48]. Many studies have shown that the level of CK is
comparatively higher in skeletal muscle than in other muscles as this muscle exhibit more contractile activities [49]. However, differences in the level of CK are found among the types of muscle fibres that individuals possess. Serum enzyme creatine kinase activity is more in the individuals who possess a greater percentage of type II muscle fibres after strenuous muscle damaging activity [27,50]. In fact, CKMM is actually being measured, when measuring total CK or just serum CK in normal serum from healthy adults, as it accounts for the enormous majority of CK. Thus, CKMM holds a vital role for diagnosis, monitoring, and assessment in MSI [4,5].

It has been shown that reactive cysteine, Cys283, forms part of a conserved cysteine-proline-serine (CPS) motif and has a pKa of about 3 pH units below that of a regular cysteine residue in muscle creatine kinase (MCK) of human beings [51]. While, the MCK promoter is under the control of myogenic transcription factors, like MyoD-I, and thus parallels the upregulation of other muscle-specific protein isoforms [27,52].

5. Variations of serum enzyme CK

However, studies have also shown the variations of serum CK levels among general populations, along with different stages of life. Base levels of serum enzyme CK are variable 35–175 U/L [53] with ranges from 20 to 16,000 U/L [17]. The levels are found higher among infants than in adults, with values that reach adult levels within the first 10 days of life [54]. While in women, the level decreases during pregnancy and increases around late gestational stage [55].

Accordingly, CK level is found to decline with the age [12]. Similarly, research also has shown the variations among male and female. Either at rest or after muscular exercise, the serum CK is found lower among female than in male, which is concluded to be due to the oestrogen hormone in a female that limits CK leak from the damaged cells [14]. Even CK levels are higher amid black males, however, some studies show no differences in the CK serum values among black and white athletes [13]. Nevertheless, CK level is found to vary according to age, gender, race, body mass and physical activity [10,11].

6. Significance of elevated serum enzyme CK

The normal values of CK level range from 52 to 336 units per L, or U/L for adult males, while 38 to 176 U/L for adult females [56]. When the CK level elevates more than the normal, it usually indicates a serious stress or disturbance to muscle tissue. The level increases significantly, depending upon muscle damage severity [45]. And various reasons can damage skeletal muscles.

Therefore, the elevation in the levels of serum enzyme CK depends on sarcomeric damage arising either from the physiologic activity like strenuous exercise or from muscular pathologies. During the process of muscle degeneration, muscle cells rupture and their contents within leak into the bloodstream. Since most of the CK in the body normally exists in muscle, elevation of serum enzyme CK in the blood signifies that muscle damage has occurred, or is occurring. Accurate history and a correct diagnostic approach with the aid of this biomarker may be of great help to formulate the correct diagnosis [10].

6.1. Pathological CK elevation in skeletal muscle

Though the primary skeletal muscle injury triggers with pain, weakness, and fatigue, it is always accompanied with the serum CK elevation. The pathological conditions like spinal muscular atrophy, amyotrophic lateral sclerosis, myotonic dystrophy, muscle damage and necrosis, all show the increased level of CK-BB [57–59].

The increased CK has been useful as prognostic and diagnostic criteria in many conditions like compartment syndrome, crush syndrome, infective rhabdomyolysis, statin myopathy [60–62]. For instance; during surgery, local muscle tissue damage occurs, with CK levels significantly higher in major surgery than in minor procedures [63]. Similarly, an increment of CK levels has been observed following convulsive seizures and heat strokes that can lead to rhabdomyolysis, a type of MSI [10]. Thus, monitoring serum enzyme CK level has been a reliable biomarker in pathological muscle injury conditions.

6.2. Physiological CK elevation in skeletal muscle

Normally, during and after exercise, the muscle fibres tear and repair subsequently. And during the process of muscle adaptations, serum CK releases in response to muscle damage. But when intense exercise leads muscle to exceed the loading limits, it harms the skeletal cellular structure at sarcolemma and Z-disks level which increases the membrane permeability, allowing CK to leak into the interstitial fluid, then enters circulation via the lymphatic system. As a result, significant serum CK elevates inducing musculoskeletal injury [64,65].
Many studies have suggested the elevation of CK levels after prolonged strenuous exercise accompanied with musculoskeletal injury. For instance; Kenney K, et al. [66] researched among 499 recruited soldiers undergoing basic training for 2 weeks, which suggested that CK levels increased greater than 50 times the upper limit of normal with muscle pain [66]. The eccentric muscular contractions containing marathon running, weight-bearing exercise and downhill running activities persistently elevate serum CK levels [10]. And the similar results were also found in the study among 46 highly trained athletes conducted by Anugweje, K.C. and Okonko, I.O. [67]. CK levels are also distinctly increased in the pre-clinical stages of muscle diseases [10] which would be useful as a tool for early detection of musculoskeletal diseases.

Moreover, the conceptualization of regular physical activities with low or moderate intensity has been recommended for good health progress and a social trend towards regular fitness has grown immensely, which is a good sign. However, beginners often experience fatigue and injury with daily exercise. In this context, serum enzyme CK could be an objective biomarker for fatigue and overtrain, as there is a lack of definitive biomarker to predict fatigue and overtraining [39].

7. Discussion

The purpose of this review paper mainly focuses on presenting the significance of elevated serum enzyme CK as a biomarker for MSI and if the elevated concentrations are helpful for early detection of MSI. CK, being a central controller of cellular energy homeostasis forms a large pool of rapidly diffusing phosphocreatine for temporal and spatial buffering of ATP levels, interchanging creatine into phosphocreatine reversibly [15]. It is predominantly found in large energy demanding tissues like skeletal muscle. To a greater extent, many studies showed predominance elevations of CK serum levels relevant to musculoskeletal injury (MSI). And plays a vital role as a marker of the functional status of muscle tissue both pathologically and physiologically [10,67]. An increment in these enzymes may indicate cellular necrosis and tissue damage following acute and chronic muscle injuries [67,68].

Even the study of CK levels has been very effective for obtaining information about sports-related musculoskeletal injuries [10]. The physical status of healthy individuals may correlate its training with the higher levels of serum enzyme CK, while if it persists during rest, it indicates the sign of subclinical muscle disease so that appropriate counseling for undertaking physical activity at lower intensity might be preventable for further muscle injuries and allow ample recovery session to athletes [10,67].

Nonetheless, the serum levels vary exceptionally among general populations in the baselines. Total CK levels depend on age, gender, race, muscle mass, physical activity and climatic condition [67]. It has made difficult to implement the acceptable normal values of CK levels without bias. The study conducted by Black HR et al. [69] showed mean total CK was 147 U/L (range of 7 to 284 U/L) for 57 black males, 61 U/L (range 35 to 87 U/L) for 44 white males, 66 U/L (range 16 to 116 U/L) for 90 black females, and 37 U/L (range 19 to 55 U/L) for 99 white females. This issue highlights a lack of specificity when laboratory reference values for serum enzyme CK do not consider other important factors like muscle mass and race [4].

Also, this unclear consensus exists on what threshold of CK elevation compares with clinically relevant MSI. Though the normal reference ranges have been implemented, some studies showed contrast effect beyond these ranges [70]. A relationship between CK elevation and the severity of the injury has been established (>6000 IU/L), but patients can have a significant injury with only moderately elevated CK levels [71]. Similarly, changes in the enzymes were found among normal healthy athletes, which in fact do not always suggest the skeletal muscle injury [10]. These factors somewhat show limitations of serum CK as a marker for muscle damage. It would be better to reset the acceptable lower and upper limits of normal CK levels and establish new standard ranges that would enable greater scientific reliability [17].

Similarly, one study done by J. Friden et al. [72] revealed that serum creatine kinase levels provided poor predictive power in estimating skeletal muscle function after eccentric exercise. Though the study was conducted in the rabbit model, it has already been demonstrated that the human muscle activations and rabbit muscle activations are maximally activated during the eccentric exercises. Since there are serum elevations but here the authors show concerns about the absolute values of the serum creatine kinase levels not providing enough information regarding the extent of the injury [72]. These results could somewhat impose limitations to CK enzymes as a marker for muscle injury. As such, some researchers have moved from associating CK with ‘muscle damage’ to ‘membrane damage’ or ‘membrane permeability’ [73,74].
It is suggested here that though there prevail some limitations, one fact remains common among all these researchers that there is an elevation in the serum CK enzyme leading to MSI. There is no doubt regarding the relationship of elevated serum and the MSI. But the serious concern about what extent of elevation is specific for what level of injury is a prerequisite to reveal. Mostly, the appearance of CK serum following low- to moderate-intensity exercise as well as at rest condition is lacking the definitive conclusion. Some authors found total serum CK elevations for 24 h after the strenuous exercise and gradually return to basal levels when at rest, while other demonstrated high levels of CK at rest also [10]. For this, further experiments and researches should be performed following detailed parameters and guidelines protocols.

8. Conclusion

To conclude, elevation of CK enzyme has relative predominance evidence with the skeletal muscle injury. It is relevant to use CK for early detection Of MSI. However, diagnostic challenges normally occurring due to isolated rise in serum CK levels or unexplained elevations in a healthy individual. For this, relevant parameters or other alternatives should be explained. Furthermore, addressing the conflicts relating its sensitivity and not establishing the extent of the injury are also useful for future perspectives. Otherwise, serum enzyme CK holds the significant role as the potential biomarker for detecting musculoskeletal injury (MSI).

9. References


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