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CREATINE KINASE RESPONSE TO HIGH-INTENSITY, SHORT-DURATION EXERCISE

BY

© MURRAY G. B. ALLEN

A THESIS

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submitted by MURRAY G. B. ALLEN

in partial fulfillment of the requirements for the degree of MASTER OF SCIENCE.

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ABSTRACT

15 healthy male oarsmen from the Edmonton Rowing Club volunteered to participate in a 90 second maximal anaerobic power test. This study was designed to examine the possibility of exercise of a high-intensity and short-duration eliciting creatine kinase (CK) activities comparable to those reported following exercise of a prolonged duration (ie: marathon). Mean delta (Δ) total creatine kinase (TCK) activity increased significantly ($p<0.01$) at both the 5 minute post (25.98 %) and 24 hour post (85.13 %) exercise sampling period. The predominate CK isoform found in the 24 hour post samples was the skeletal muscle specific isoenzyme, CK-MM. Mean CK-MB content in the serum at the 24 hour post period showed an absolute value of 3.12 ± 0.19 (IU/L) which was calculated to be 4.46 % of the 24 hour TCK measure. A very low degree of commonality (correlation range, 0.002 to 0.261) was demonstrated between the ΔTCK values and performance measures ($\dot{V}o_{2\ max}$ ($ml \cdot kg^{-1} \cdot min^{-1}$) and 90 second power output (watts)). In conclusion, the high-intensity, short-duration bout of exercise elicited significant increases in the CK activity. Nevertheless, these changes were not of the same magnitude as those witnessed on marathon completion. Furthermore, it is suggested that the reduced duration of the exercise bout, predominant contraction type employed and assaying conditions utilized in the experimental procedure were strongly implicated in the characterization of these findings.

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Chapter I

INTRODUCTION

Creatine Kinase (CK) and its isoenzymes have been studied extensively as biochemical markers used in the diagnosis of myocardial and skeletal muscle disorders (Rogers et al., 1985; Gunderson et al., 1983). Although serum enzyme levels are used frequently as a measure of cell injury, it is not always clear what their release really signifies with respect to the extent of damage or survival of the cell (Skillen, 1984). The phrase "cell injury", is used to describe a wide variety of cellular conditions, ranging from a transient departure from optimal functioning, with no long-term adverse consequences, to severe irreversible damage leading to the death of the cell (Danpure, 1984).

The effect of exercise on serum enzymes has been well documented, nevertheless, there appears to be considerable controversy and inconsistency in the reported findings. Physical exercise has been demonstrated to elicit increased activity levels in several serum enzymes. This exercise-induced release of muscle enzymes into the circulation has been related to both the intensity and duration of the exercise period. Evidence to date shows great controversy surrounding the possible dominate factor (intensity or duration) in promoting these augmented serum levels. Fowler et al., (1968); Hunter et al., (1971); Chahine et al., (1976) and Lott & Landesman, (1984) suggest that the duration of the activity is of fundamental significances in the modulation of the exercise induced serum enzyme response. Berg (1978); Galteau et al., (1976) and

Tiddus et al., (1983) contend that the intensity with which the exercise is performed is the predominate factor underlying the serum enzyme fluctuations.

The majority of the work done in this area of late has dealt with the serum enzyme response to exercise of a prolonged nature (ie. marathons and triathlons)(Apple et al., 1987; Apple & Rogers, 1986; Apple et al., 1985; Jansson & Slyven, 1985; Noakes et al., 1983; Rumley et al., 1985; Rogers et al., 1985). Furthermore, very little research has examined the area of serum enzyme responses to high intensity, short duration bouts of exercise.

It is hypothesized that the intensity with which an exercise is performed is an important contributing factor in the enhanced total creatine kinase (TCK) response witnessed post exercise. Thus, the principal aim of the present study was to determine if exercise of short duration and high intensity could elicit alterations in the serum TCK activity comparable to those reported following exercises of a prolonged duration (ie. marathon). A rowing exercise which employs large muscle groups at a high intensity (ie. 90 second maximal anaerobic power test) was considered an ideal means by which enzymatic responses could be elicited.

On completion of long duration types of exercise the CK isoenzyme profiles have shown augmented levels of heart specific CK-MB isoforms in some individuals. These findings have not been documented in activities other than those of a prolonged nature. Therefore, if elevated TCK levels are produced in the present experiment, it would be desirable to elucidate the specific CK

isoenzyme content present in the serum following the acute exercise interval and to provide an indication as to their source.

Chapter II

METHODS

Prior to the present study, a pilot experiment was performed in an attempt to observe whether short duration exercise could elicit significant increases in measured TCK activity. Five sedentary individuals participated in a 45 second fatigue test on the Cybex 2 Isokinetic Dynamometer. The results of that experiment showed a very interesting response in that an increase of 46.7 % was witnessed in the groups mean 24 hour post TCK activity. Moreover, one particular individual injured a hamstring muscle while performing the test. His pre, 5 minute post and 24 hour post serum TCK can be viewed in Figure 1. An increase of 1054 % was witnessed on the 24 hours post TCK measure. This individuals TCK activities were not included in the group means described above. These findings formed the basis for a detailed study in this area of exercise-induced alteration in muscle enzymology.

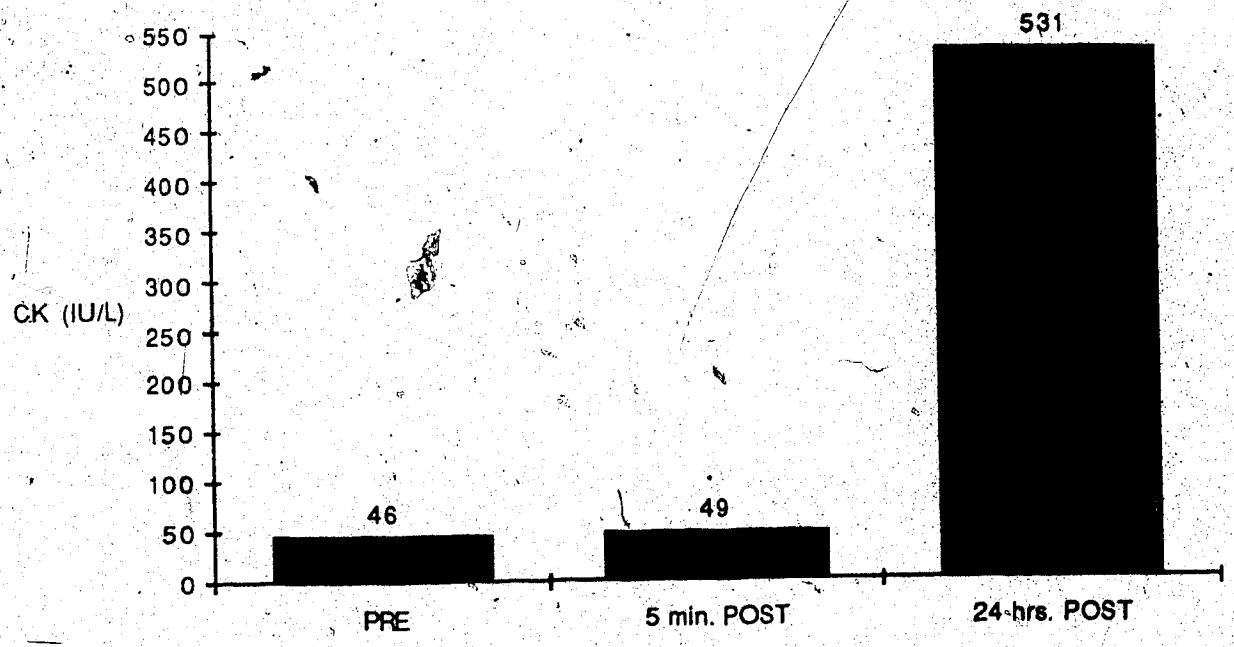
Fifteen trained oarsmen consented to participate in a 90 second maximal anaerobic power test, which required the athletes to complete as many revolutions possible in 90 seconds at resistance of 3 kp on a rowing ergometer (Gjessing Ergorow). The subjects characteristics are present in Table 1. Subjects were asked to refrain from physical activity for two days prior to testing.

Venous blood samples were collected by a certified laboratory technician from the anterior cubital vein prior to testing, 5 minutes post and 24 hours post exercise. All samples were allowed to clot followed by immediate centrification which in turn was followed by

sample storage at -70 degrees Celsius pending analysis. TCK and CK-MB activity was determined spectrophotometrically as per Sigma chemical technical Bulletin #45-UV and BMC Immunoinhibition Bulletin #300691 respectively. All samples were analyzed in duplicate by the researcher and the University of Alberta Hospital to insure value reproducibility. Samples were found to be within ± 5 %.

Figure 1:

ΔTCK Activity (IU/L) Pre, 5 min. Post & 24 Hrs. Post
45 sec. Cybex Fatigue test



$\dot{V}o_{2\ max}$ was assessed three days prior to the 90 second power test. This measure was determined during an incremental test on the aforementioned ergometer. Resistance was increased 25 watts/min. for seven minutes at which time the subjects were instructed to give an all out maximal effort for the last two workloads.

To confirm that $\dot{V}o_{2\ max}$ was obtained, all subjects met at least one of the following criteria; oxygen consumption peaked and/or plateaued, age predicted maximal heart rate was attained, an R value of greater than 1.10 was reached and/or volitional exhaustion occurred. Expiratory gases were collected and analyzed every thirty seconds via a Beckman Metabolic Cart. Gases of a known concentration were used for calibration before and after each test. Heart rate was recorded each minute with a PE 3000 Sport Tester heart rate monitor.

At the time of testing the subjects were involved in pre-season training program which consisted of exercising five days a week on a Concept 2 rowing ergometer, at an intensity equal to the heart rate at 75% of $\dot{V}o_{2\ max}$. Each training session consisted of a minimum of 40 minutes of continuous rowing.

Paired t-tests were employed to determine significance between the pre, 5 minute post and 24 hour post exercise TCK activities. A Pearson Product Moment Correlation was computed between the following variables; $\dot{V}o_{2\ max}$, 90 second power output, ΔTCK pre to 5 minutes post exercise, ΔTCK pre to 24 hours post exercise and ΔTCK 5 minute post to 24 hours post exercise.

Table 1.

Physical Characteristics of the Subjects

Subject	Age (years)	Height (cm)	Weight (kg)	$\dot{V}O_{2\ max}$ ($ml \cdot kg^{-1} \cdot min^{-1}$)	Power Output 90 Seconds (Watts)
1	23	181.9	89.7	57.5	453.0
2	28	171.2	67.8	59.1	347.0
3	21	169.3	79.8	60.1	345.2
4	20	188.2	82.7	53.2	443.9
5	21	185.3	81.2	60.8	413.5
6	18	184.8	66.4	62.9	363.6
7	23	182.1	79.5	59.8	388.4
8	29	192.1	87.1	53	424.7
9	21	193.1	84.1	54.6	456.9
10	28	189.7	94.2	48.1	433.6
11	21	185.4	78.9	59.1	415.4
12	25	184.3	85.7	53.3	401.8
13	20	188.9	69.2	58.9	358.2
14	22	173.8	72.8	58.2	400.0
15	22	179.5	72.4	59.4	390.6
MEAN	22.8	183.3	79.4	57.2	402.4
SEM	0.8	1.8	2.1	1.0	9.6

Chapter III

RESULTS

The effects of the 90 second bout of exercise on the measured TCK response are shown on Table 2. The mean TCK activity was demonstrated to increase significantly ($p < 0.01$) following the 90 second exercise bout at both the 5 minute and 24 hour post exercise time period .

Table 2.

Mean Relative and Absolute TCK Activity Prior to, 5 minute and 24 hours Post Testing.

	PRE	5 min. POST	24 hrs. POST
ABSOLUTE TCK (IU/L)	37.83	47.61 [a]	69.98 [b]
% TCK RELATIVE to PRE SAMPLE	100.00 %	125.98 %	185.13 %

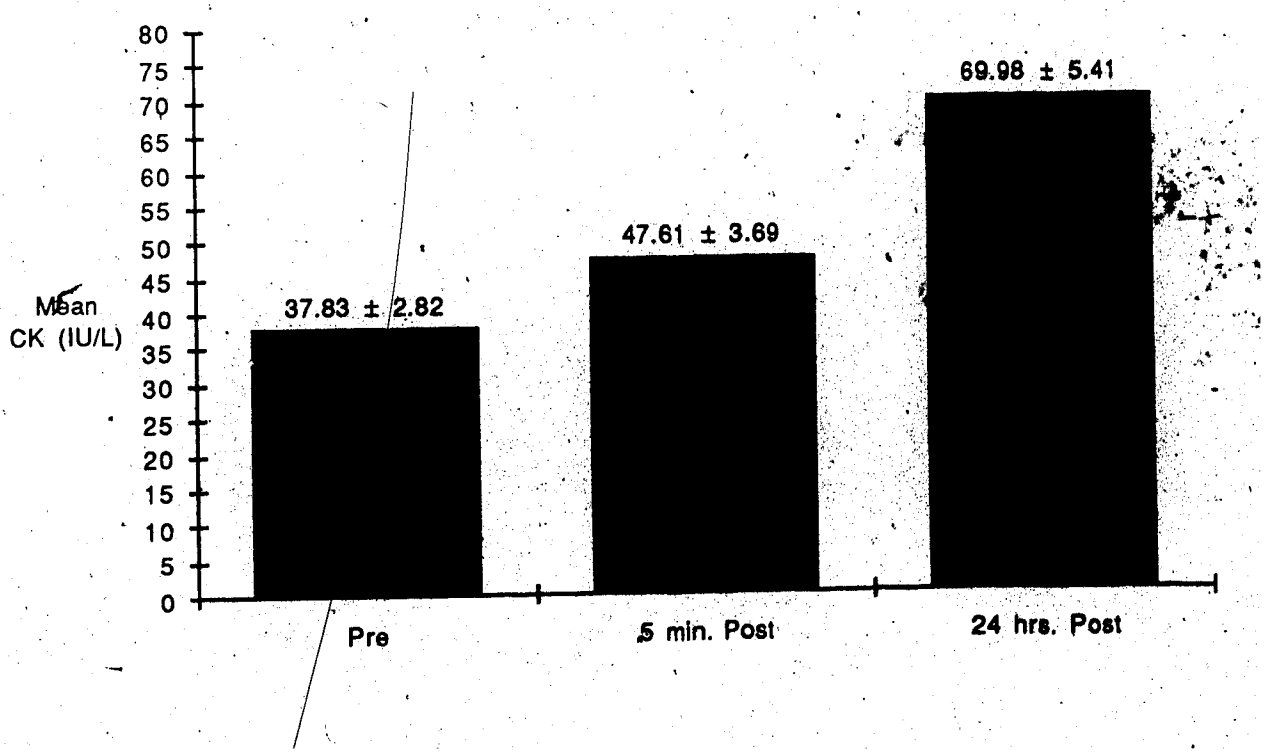
a = significantly different from pre test sample, $p < 0.01$.

b = significantly different from pre and 5 min. post sample, $p < 0.01$.

An increase in TCK activity of 26 % was witnessed 5 minutes following the exercise bout while the 24 hour TCK response showed an increase of 85.13 % from the resting level (Figure 2). Mean CK-MB activities 24 hours post testing showed an absolute value of 3.12 IU/L ± 0.19 which was calculated to be 4.46 % of the 24 hour TCK measure. Apple et al. (1984) reported CK-MB activities as high as 12% of the TCK following marathons. Under non-pathophysiological conditions, normal CK-MB isoenzyme composition in the serum range from trace amounts to 3% of the absolute TCK value (Siegel et al., 1983).

Figure 2.

Mean TCK (IU/L) Activity; Pre, 5 minutes Post and 24 hours Post Exercise



The degree of correlation between the measured variables are shown in Figures 3 through 8. These values indicate a low degree of commonality between the variables examined. The 90 second power outputs attained in the present study ranged from 345.2 to 456.9 watts. These power outputs demonstrated very little correlation (correlation range, .002 to .261) with the ΔTCK activities recorded. Furthermore, $\dot{V}o_{2\ max}$ measures (range 48.1 to 62.9 ($ml \cdot kg^{-1} \cdot min^{-1}$)) showed next to none, commonality (correlation range, .001 to .107) with the measured ΔTCK responses.

Figure 3:

ΔTCK Activity Pre to 5 min. Post (IU/L)
As Compared to 90 sec. Power Output (Watts)

r= 0.011
n= 15

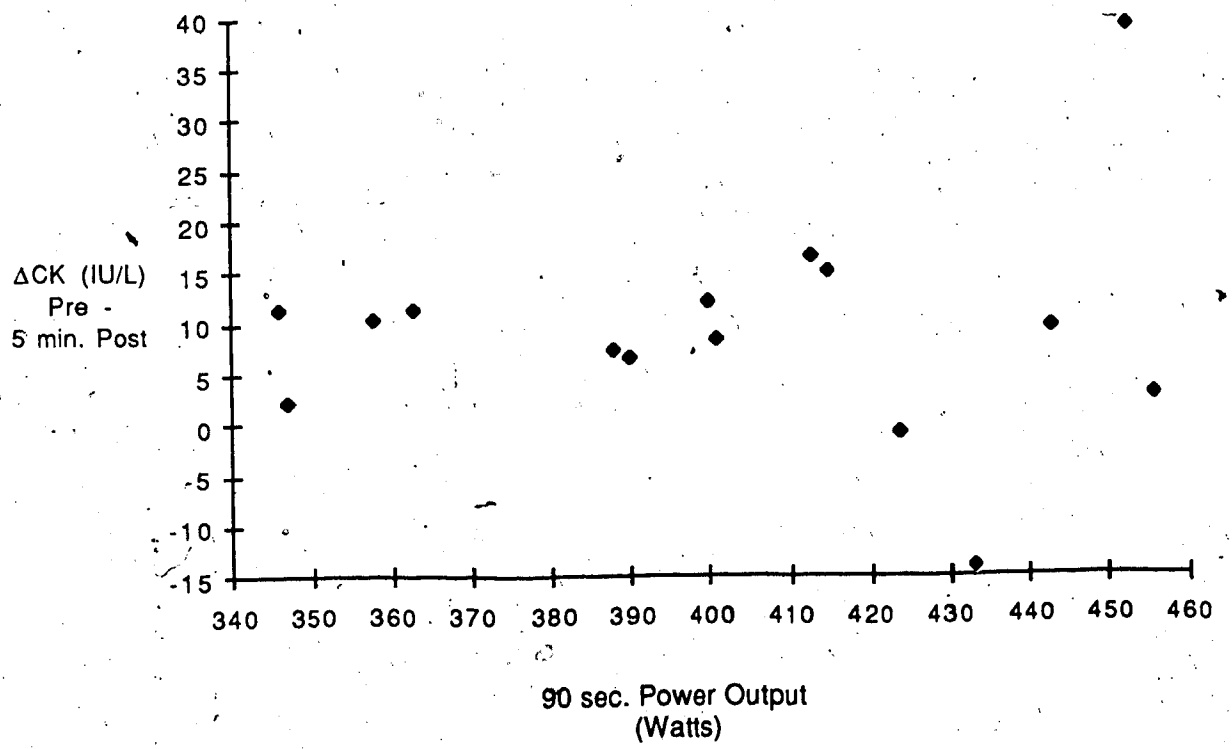


Figure 4:

ΔTCK Activity Pre to 24 hrs. Post (IU/L)
As Compared to 90 sec. Power Output (Watts)

r= 0.107
n= 15

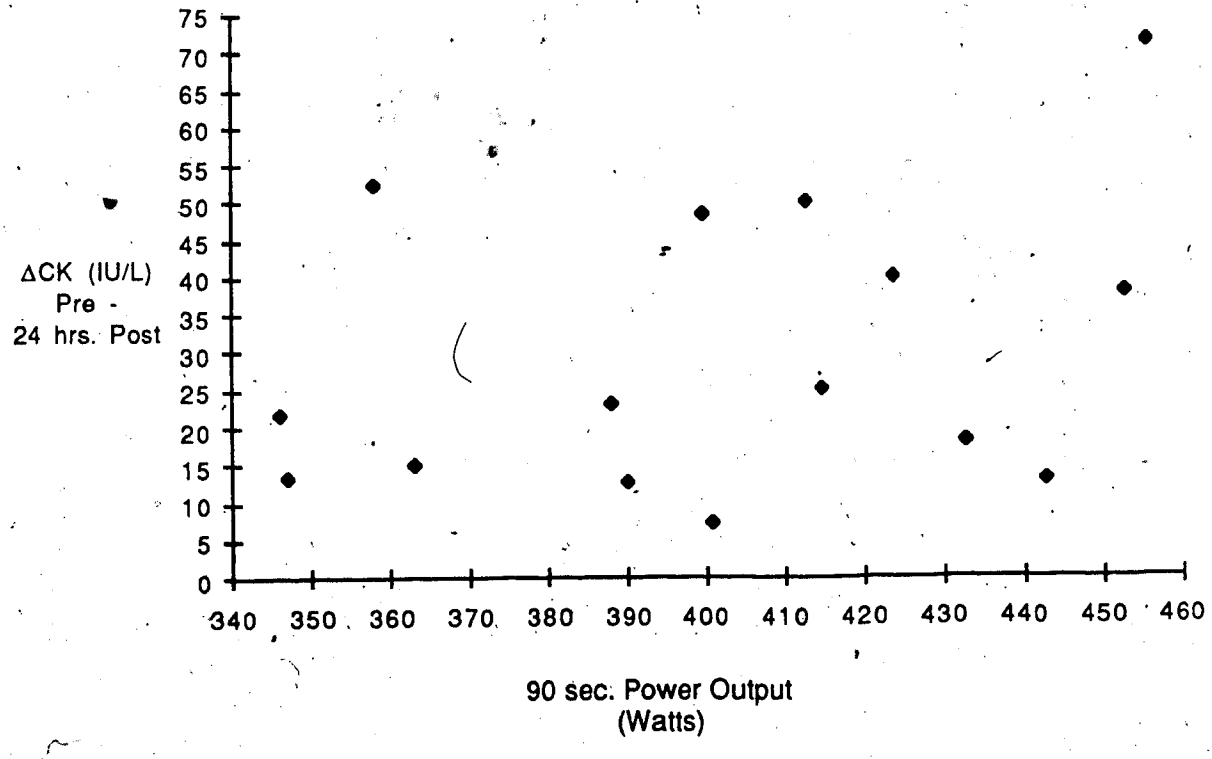


Figure 5:

ΔTCK Activity 5 min. Post to 24 hrs. Post (IU/L)
As Compared to 90 sec. Power Output (Watts)

r= 0.057
n= 15

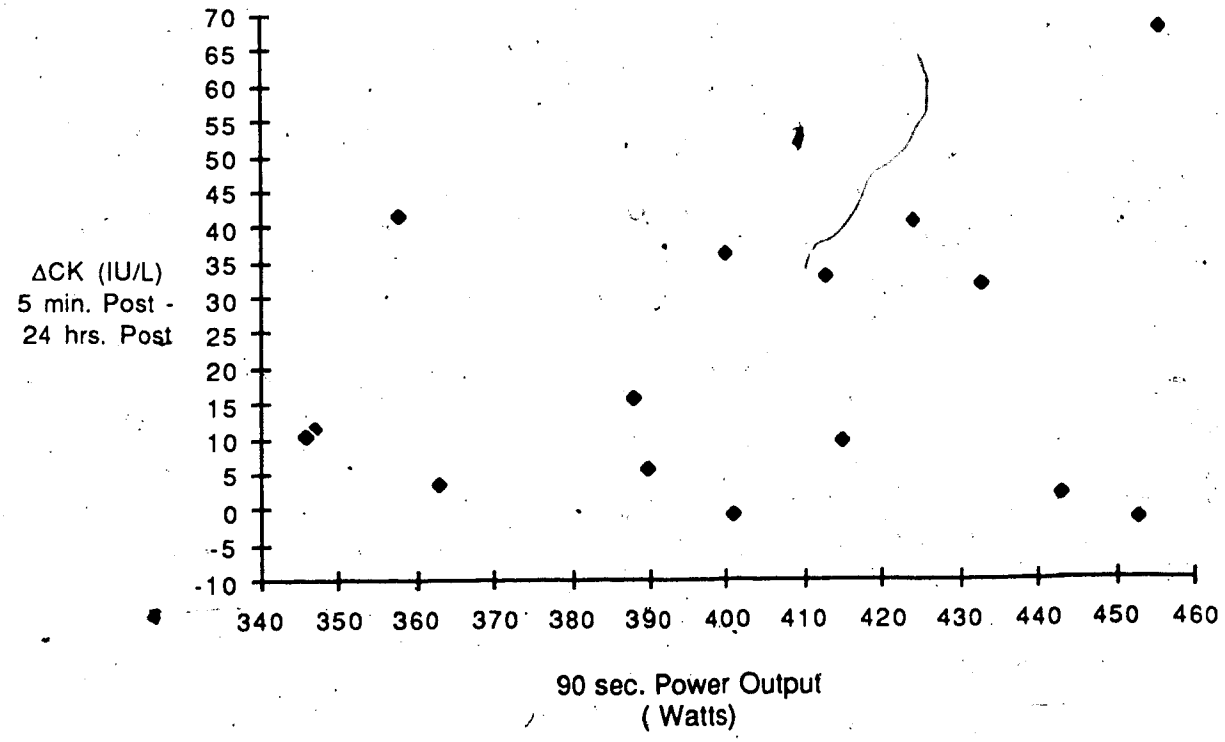


Figure 6:

ΔTCK Activity Pre to 5 min. Post (IU/L) As Compared to $\dot{V}O_{2\ max}$ ($ml \cdot kg^{-1} \cdot min^{-1}$)

r= 0.261
n= 15

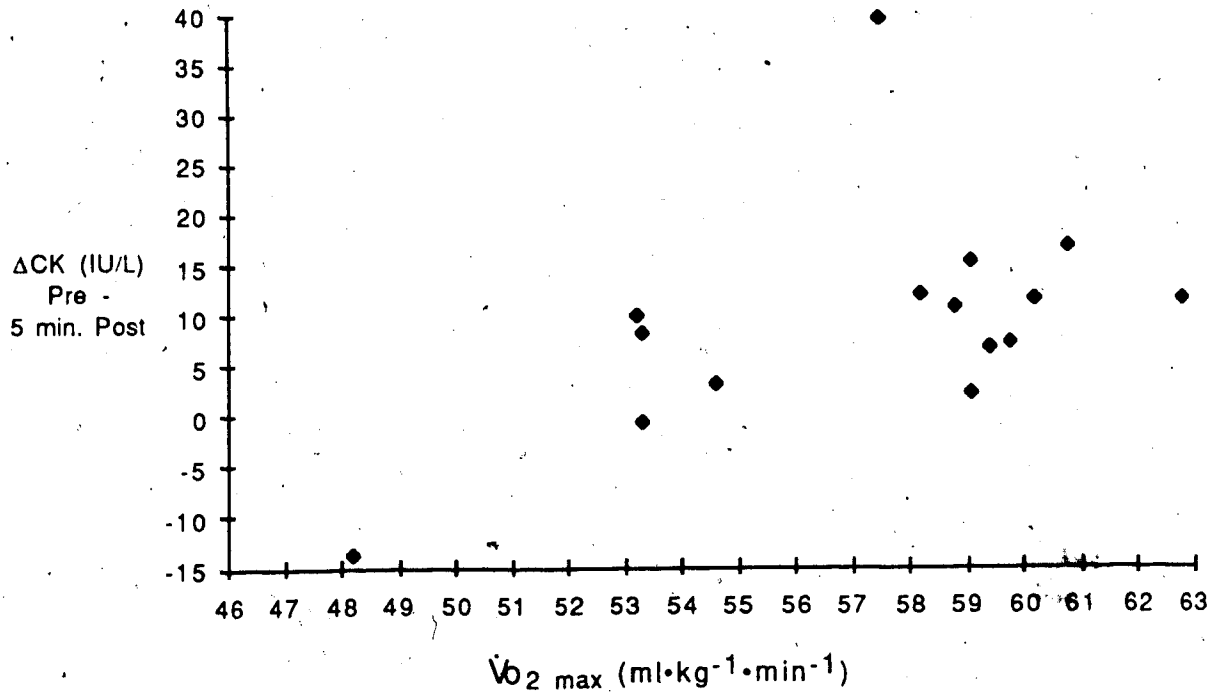


Figure 7:

ΔTCK Activity Pre to 24 hrs. Post (IU/L) As Compared to $\dot{V}o_{2\ max}$ ($ml \cdot kg^{-1} \cdot min^{-1}$)

r= 0.002
n= 15

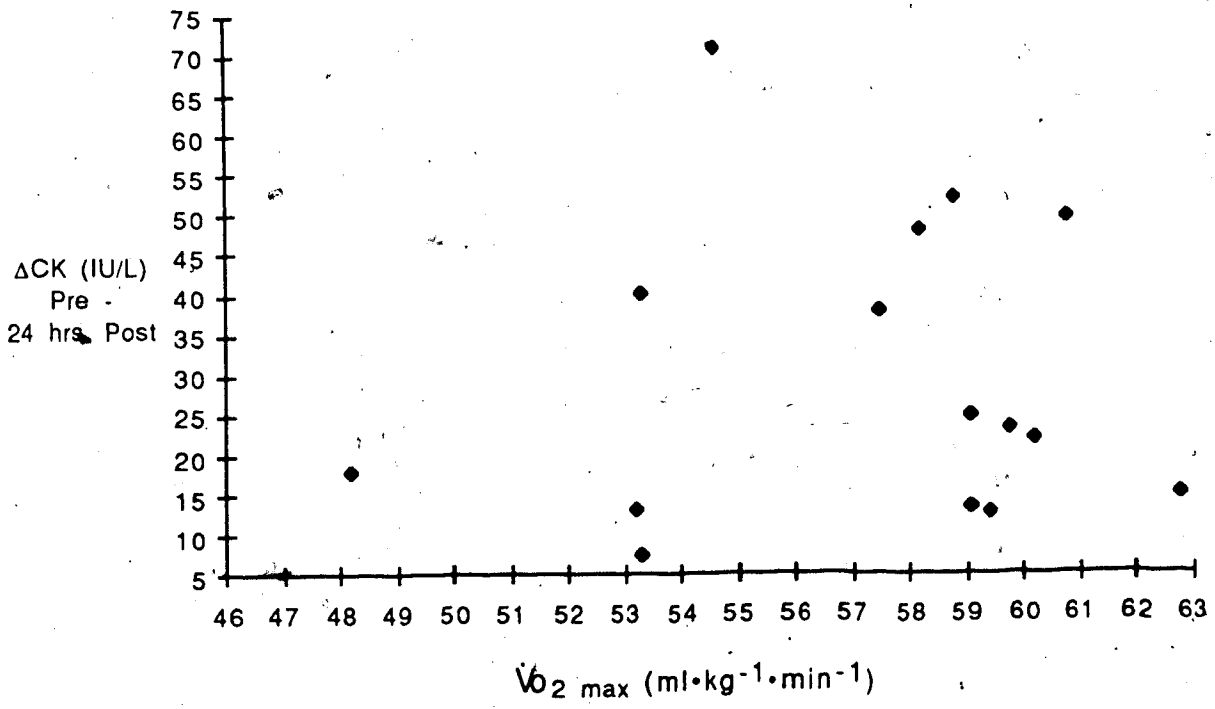
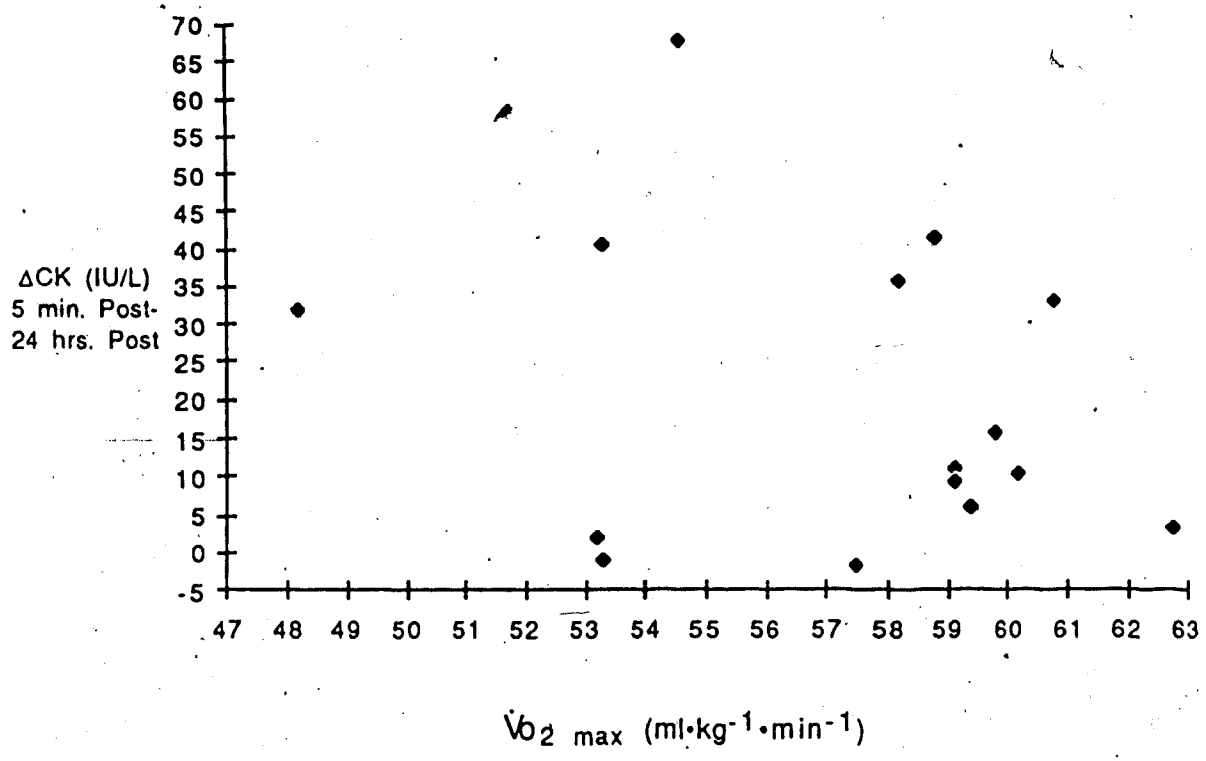


Figure 8:

ΔTCK Activity 5 min. Post to 24 hrs. Post (IU/L) As Compared to $\dot{V}O_{2\ max}$ ($ml \cdot kg^{-1} \cdot min^{-1}$)

r= 0.057
n= 15



Chapter IV
DISCUSSION

Total creatine kinase activity following the 90 second bout of maximal exercise showed significant increases at both the 5 minute and 24 hour post sampling time (Table 2). The TCK response appears to be a temporally dependent event, whereby, maximal (peak) values are said to be obtained anywhere from 8 to 24 hours post exercise (Rogers et al., 1987; Apple et al., 1987; Priest et al., 1982). It is difficult to pin point the peak response without exhaustive serial sampling. Consequently, based on previous research, the 24 hour sample was taken to represent the peak TCK activity attained. Evans and co-workers (1986) have demonstrated that myofibrillar damage is greater 3 days following a strenuous exercise bout than immediately after. This would indicate that the degeneration of the contractile unit is a delayed event that coincides with the enhanced TCK activity viewed post exercise. The resting TCK levels (Table 2) of the subjects utilized in the present study are close to the upper range for normal basal TCK activities (8-50 IU/L)(Bias & Edwards, 1982). These elevated resting measures could be attributed to the subjects enhanced muscle mass (Norton et al., 1985) and/or the augmented protein turnover witnessed in trained athletes at rest (Priest et al., 1982). On isoenzyme analysis it was concluded that the TCK cleared in the current study was composed principally of the CK-MM isoform. This isoenzyme is predominantly found in the skeletal tissue (Nicholson et al., 1985). CK-MB values of greater than 6% of the TCK are said to be characteristic of acute myocardial

Periodically, CK-MB activities of this magnitude are reported post marathon (Apple et al., 1987; Apple & Rogers, 1986; Apple et al., 1985; Jansson & Slyven, 1985; Noakes et al., 1983; Rumley et al., 1985; Rogers et al., 1985). However, none of the subjects employed in the current study had values of this order.

The TCK activities obtained in the present study show a similar clearance pattern to those witnessed post marathon, however, the magnitude of the response is reduced with regard to peak TCK levels attained. This reduction could be attributed to primarily four factors: (a) exercise conditions (ie. intensity and duration); (b) subjects level of training; (c) predominate contraction type employed in the exercise bout; (d) laboratory assaying conditions.

(a) Since the exercise duration utilized in this study was quite short (ie. 45 or 90 sec.), it probably played a significant role in contributing to the lower than expected post exercise TCK values. Although the TCK levels did not approach those reported post marathon (Apple et al., 1987; Apple & Rogers, 1986; Apple et al., 1985; Jansson & Slyven, 1985; Noakes et al., 1983; Rumley et al., 1985; Rogers et al., 1985), the 85% increase noted in TCK activity at the 24 hour post exercise time period would suggest a role for exercise intensity in the manifestation of the enhanced TCK clearance observed post exercise. The precise contribution of exercise intensity and/or duration can not be elucidated from the results of this study; however, the importance of standardizing exercise intensities or work loads when comparing individual TCK results is of extreme importance.

(b) The subjects degree of physical conditioning has been demonstrated to play a significant role in the characterization of the TCK response (Apple et al., 1987; Jansson et al., 1985; Brown, 1988). Nevertheless, the results of the present study showed no relation between TCK activity and subjects physical condition as measured by $\dot{V}o_{2\ max}$ and 90 second power outputs (Figure 3-8). Perhaps, this lack of correlation evidence may be due to the small n employed or the limited spread of data found in the present study. TCK content has been shown to be higher in type II fibers than in type I. With the subjects working at the intensity utilized in the present study, the majority of the muscle fibers recruited should theoretically be of a type II nature. The increase in TCK activity post testing is consistent with this notion, whereby, it is speculated that the type II fibers are predominantly recruited during the 90 second exercise bout and consequently are most likely the sites of cellular damage and TCK clearance. When an individual trains for specific contractile activity a decrease in delayed muscle soreness and enzymatic streaming is witnessed (Armstrong et al., 1984). The athletes employed in the current study were trained and tested on virtually identical pieces of equipment. Perhaps the subjects habituation to the contraction types and sequencing contributed to the lower than expected TCK activities. Newham et al, (1986) states that the extent to which the active muscle is lengthened and the degree of habituation to eccentric work plays an significant role in determining the extent of the enzyme response.

(c) The mechanical pounding and eccentric components of running are thought to contribute to the increase in TGK activity witnessed post exercise. Gunderson and co-workers (1983) suggest that distance running may cause a state of cell membrane hyperpermeability in the skeletal muscle which in turn facilitates enhanced cytoplasmic content streaming into the circulation. The increased membrane permeability viewed by the elevated serum enzyme levels indicates that other crucial cellular components may likewise efflux into the serum. Under these circumstances cellular function would probably be retarded. Furthermore, the ability of the cell to sustain muscular contraction would be reduced. Fowler et al. (1968) found no clear relationship between the molecular weight of the enzyme and the degree to which the enzymes appeared in the serum. Rowing, however, is characterized by not only an absence of such localized trauma but also by reduced force production. Forces of 130 - 275 kg have been measured during the foot strike phase of running (Symanski et al., 1983). Secher (1983), contends that the magnitude of the force required to develop dynamic strength in rowing is equal to 84 kg. Both the energy demands and forces incurred by the oarsman are somewhat different than those placed on athletes of other exercise modes. Rowing utilizes the muscles of the arms, back and legs, in a concentric manner, while activities such as running are almost entirely dependent on the muscles of the legs in both an eccentric and concentric fashion. Moreover, rowing is a weight supported activity in contrast to a non weight-supported activities such as marathon running. Consequently, in the present study the predominate contraction type employed during the activity

may have contributed to the reduced TCK activity witnessed post exercise.

(d) As mentioned previously the TCK activities recorded in the present study were significantly elevated, although, it was not at all comparable to the magnitude of the results viewed post marathon. It is critical to note that any biochemical measure is assay dependent in that the pH, temperature and buffer mediums utilized in the procedure have a powerful influence on the viewed results. Although temperature correction factors were utilized in the assaying procedure employed in the present study, some variance in TCK activity after temperature correction was observed. Thus, the sensitivity of the assaying medium temperature on the TCK response plays a critical role in the viewed results. Keeping this in mind it is therefore difficult to compare the results of the current study to those viewed by other researchers.

The clearance of TCK and its isoenzymes into the circulation is thought to be facilitated by two mechanisms which both effect sarcolemma permeability. These being mechanical and/or metabolic factors. The mechanical effect of exercise that requires contraction types of an eccentric nature have been shown to alter the muscle ultra structure (Newham et al., 1986; Jansson et al 1985; Armstrong 1984). Eccentric contractions produce a higher tension per active motor unit than either concentric or isometric contractions, yet the metabolic cost in producing such forces is less than that seen with isometric or concentric contractions. It is speculated that the great tension produced by eccentrically contracting fibers ultimately facilitates the enhanced TCK response

seen post exercise due to the damage incurred to the contractile unit during the exercise bout.

The metabolic depletion of ATP in exercising tissues may be responsible for the enhanced TCK activity witnessed post exercise (King et al., 1976; Cheetman et al., 1986; Maxwell et al.,1981). The argument presented in the literature is that with exhaustive exercise a situation exists in the working tissues in which cellular processes are in a sense competing for the ATP that is normally allocated to the maintenance of cell membrane integrity. A reduction in the sarcolemmic integrity increases the cells permeability which in turn facilitates the enhanced clearance of CK from the exercising tissue. Perhaps the exercise bout employed in the current study was not of a long enough duration to deplete the ATP levels sufficiently to allow for a rise in the TCK activity to a level similar to those witnessed following marathons. However, the intense nature of this activity would not allow for ATP production to be maintained. A fall in ATP levels most likely accompanied this exercise condition and therefore, may have facilitated the increase in TCK activity. Obviously this requires further investigation before the role of ATP depletion can be implicated conclusively in the altered sarcolemmic permeability witnessed during intense exercise. Training may afford some degree of protection against the cell damage due to an increase in the size and number of mitochondria (Holloszy et al., 1975; Hunter & Critz, 1971). In this situation the potential for ATP production is enhanced considerably. As the level of ATP availability increases in the cell, the potential for a reduction in cellular integrity is decreased. Therefore, the mitochondrial proliferation

witnessed with aerobic training enables the tissues to work, for a greater period of time before substantial increases in TCK are observed in the serum.

In summary, it was found that exercise of a high intensity and short duration elicited significant increases in the post exercise TCK response. The predominate CK isoenzyme observed under these circumstances was the skeletal muscle form, CK-MM. The TCK values obtained in the present study were of a smaller magnitude than the post marathon data documented in the literature. It is suggested that the intensity of exercise may be a significant factor in eliciting a TCK response similar to that viewed on marathon completion. The mechanism by which the enzyme is cleared into the circulation is uncertain. Due to the reduced absolute forces incurred in rowing and the predominate contraction types employed in this activity (ie. concentric), it is suggested that metabolic factors play a prominent role in facilitating the enhanced enzyme clearance. Nevertheless, further study is required to more clearly elucidate the significance of TCK clearance following high intensity, short duration bouts of exercise in relation to the delayed muscle soreness response.

Recommendations For Further Research

1) Further research is suggested to standardize CK assaying techniques, as methodology differences exist in the literature which makes value comparisons difficult.

2) It is recommended that an experimental study be implemented utilizing both anaerobically and aerobically trained individuals to address the issue of training specificity and the CK response. Detailed biochemical analysis employing both serum samples and tissue biopsies is recommended to further elucidate the relationship between the CK response and delayed muscle soreness.

References

Abraham, W. M. (1977). Factors in delayed muscle soreness. Medicine and Science in Sports, 9(1), 11-20.

Apple, F.S. & Rogers, M. A. (1986). Mitochondrial creatine kinase activityalterations in skeletal muscle during long-distance running. American Physiological Society. , 482-485.

Apple, F.S., Rogers, M. A., Casal, D. C., Sherman, W. M. & Ivy, J. L. (1986).Creatine kinase-MB isoenzyme adaptations in stressed human skeletal muscle of marathon runners. Journal of Applied Physiology. , 149-153.

Apple, F.S., Rogers, M. A., Sherman, W. M., Costill, D. L., Hagerman, F. C. & Ivy, J. L. (1984). Profile of creatine kinase isoenzymes in skeletal muscles of marathon runners. Clinical Chemistry, 30(3), 413-416.

Apple, F. S., Rogers, M. A., Casal, D. C., Lewis, nL., Ivy, J. L. & Lampe, J. W. (1987). Skeletal muscle creatine kinase MB alterations in women marathon runners. European Journal of Applied Physiology, 56, 49-52.

Armstrong, R. B. (1984). Mechanisms of exercise-induced delayed onset muscular soreness: A brief review. Medicine and Science in Sports and Exercise, 16(6), 529-538.

Berg, A. & Haralambie, G. (1978). Changes in serum creatine kinase and hexose phosphate isomerase activity with exercise duration. Eur. J. Appl. Physiol., 39, 191.

Besserman, S. P., Yang, W. C. T., Geiger, P. J. & Erickson-Viitanen, S. (1980). Intimate coupling of creatine phosphokinase and myofibrillar adenosinetriphosphatase. Biochemical and Biophysical Research Communications, 96(3), 1414-1420.

Bessman, S. P. & Fonyo, A. (1966). The possible role of the mitochondrial bound creatine kinase in regulation of mitochondrial respiration. Biochemical and Biophysical Research Communications, 22(5), 597-601.

Bessman, S. P. & Geiger, P. J. (1981). Transport of energy in muscle: The phosphorylcreatine shuttle. Science, 211, 448-452.

Bias, R. & Deamer, D. (1984). Creatine Kinase. CRC Clin Lab Sci, 3, 291-335.

Bloor, C. M. (1978). Effects of exercise on enzyme interpretation. The Western Journal of Medicine, 128(1), 46-47.

Bornheimer, J.F. & Lau, F. Y. K. (1981). Effects of treadmill exercise on total and myocardial creatine phosphokinase. Chest, 80(2), 146-148.

Byrnes, W. C., Clarkson, P. M., Spencer White, J, Hsieh, S. S., Frykman, P. N. & Maughan, R. J. (1985). Delayed onset muscle soreness following repeated bouts of downhill running. Journal of Applied Physiology, 710-715.

Chahine, R. A., Kazantzis, A., Luchi, R. J, Raizner, A. E & Gyorkey, F. (1976). Effects of routine treadmill testing on the serum enzymes. Cardiology, 61, 162-169.

Cheetman, M. E., Brooks, B. S. & Williams, C. (1986). Human muscle metabolism during sprint running. Journal of Applied Physiology, 54-59.

Clarkson, P. M., Litchfield, P., Graves, J, Kirwan, J & Byrnes, W. C. (1985). Serum creatine kinase activity following forearm flexion isometric exercise. European Journal of Applied Physiology, 53, 368-371.

Clarkson, P. M., Apple, F. S., Byrnes, W. C., McCormick, K, M. & Triffletti, P. (1987). Creatine kinase isoforms following isometric exercise. Muscle and Nerve, 10, 41-44.

Costill, D. L., Coyle, E. F., Fink, W. F., Lesmes, G. R. & Witsmann, F. A. (1979). Adaptations in skeletal muscle following strength training. Journal of Applied Physiology, 96-99.

Danpure, C. (1984). Lactate dehydrogenase and cell injury. Cell. Bochem. Funct., 11, 76-79.

Desmos, M. A., Gitin, E. L. & Kagen, L. J. (1974). Exercise myoglobinemia and acute exertional rhabdomyolysis. Arch. of Internal Medicine, 134, 669- 673.

Dill, D. B. & Costill, D. L. (1974). Calculation of percentage changes in volumes of blood, plasma, and red cells in dehydration. Journal of Applied Physiology, 37(2), 247-248.

Erickson-Viitanen, S., Geiger, P. J., Viitanen, P. & Bessman, S. P. (1982). Compartmentation of mitochondrial creatine phosphokinase. The Journal of Biological Chemistry, 257(23), 14405-14411.

Evans, W. J., Meredith, C. N., Canon, J. G., Dinarello, C. A., Frontera, W. R., Hughes, V. A., Jones, B. H. & Knuttgen, H. G. (1986). Metabolic changes following eccentric exercise in trained and untrained men. Journal of Applied Physiology, 1864-1868.

Fowler, M., Gardener, G. A & Kazerunian, H. (1968). The effect of exercise on serum levels. Arch. Phys. Med. Rehabil., 49, 54.

Friden, J., Sfakianos, P. N. & Hargens, A. R. (1986). Muscle soreness and intramuscular fluid pressure: Comparison between eccentric and concentric load. Journal of Applied Physiology, 2175-2179.

Friden, J. (1984). Changes in human skeletal muscle induced by long-term eccentric exercise. Cell and Tissue Research, 236, 365-372.

Galteau, M., Seist, G. & Poortmans, J. (1976). Continuous in vivo measurement of creatine kinase variations in man during exercise. Clinica Chimica Acta, 66, 89-93.

Galun, E. & Epstein, Y. (1984). Serum creatine kinase activity following a 120-km march. Clinica Chimica Acta, 143, 281-283.

Gunderson, H., Parliman, J., Parker, J. & Bell, G. (1983). Membrane permeability changes as a fatigue factor in marathon. Biochem. Exerc., 3, 877.

Hagerman, F. C. (1984). Applied physiology of rowing. Sports Medicine, 1, 303-321.

Harris, R. C., Sahlin, K. & Hultman, E. (1976). Phosphagen and lactate contents of m. quadriceps femoris of man after exercise. Journal of Applied Physiology Society, 852-857.

Hikida, R. S., Staron, R. S., Hagerman, F. C., Sherman, W. M. & Costill, D. L. (1983). Muscle fiber necrosis associated with human marathon runners. Journal of the Neurological Sciences, 59, 185-203.

Hirvonen, J., Rehunen, S. Rusko, H. & Harkonen, M. (1987). Breakdown of high-energy phosphate compounds and lactate accumulation d uring short supramaximal exercise. European Journal of Applied Physiology, 56, 253- 259.

Holloszy, J. O. (1975). Adaptation of skeletal muscle to endurance exercise. Medicine and Science in Sports, 7(3), 155-164.

Houk, T. W. & Putman, S. V. (1973). Location of the creatine phosphokinase binding site of myosin. Biochemical and Biophysical Research Communications, 55(4), 1271-1277.

Houston, M., Green, H. Thomson, J. & Reid, P. (1978). The response of oxygen consumption, body temperature, blood substrates and serum enzymes to heavy work performed over twenty-four hours. European Journal of Applied Physiology, 39, 145-154.

Hunter, J. B. & Critz, J. B. (1971). Effect of training on plasma enzyme levels in man. Journal of Applied Physiology, 31(1), 20-23.

Jansson, E. & Sylven, C. (1985). Creatine kinase MB and citrate synthase in type I and type II muscle fibres in trained and untrained men. European Journal of Applied Physiology, 54, 207-209.

Kaman, R. L. Goheen, B., Patton, R. & Raven, P. (1977). The effects of near maximum exercise on serum enzymes: The exercise profile versus the cardiac profile. Clinica Chimica Acta, 81, 145-152.

Kasperek, G. J. & Snider, R.D. (1985). Increased protein degradation after eccentric exercise. European Journal of Applied Physiology, 54, 30-34.

Kasperek, G. J. & Snider, R.D. (1985). The susceptibility to exercise-induced muscle damage increases as rats grow larger. Experientia, 41, 616-617.

Kettunen, P., Kala, R. & Rehunen, S. (1982). Creatine kinase and its isoenzymes in skeletal muscle of athletes. The Lancet, 611.

Kettunen, P., Kala, R. & Rehunen, S. (1984). CK and CK-MB in skeletal muscle of athletes and in serum after thoracic contusion in sport. J. Sports Med. 24, 21-25.

King, S. W., Statland, B. E. & Savory, J. (1976). The effect of a short burst of exercise on activity values of enzymes in sera of healthy young men. Clinica Chimica Acta, 72, 211-218.

Kirwan, J. P., Clarkson, P. M., Graves, J. E., Litchfield, P. L. & Byrnes, W. C. (1986). Levels of serum creatine kinase and myoglobin in women after two isometric exercise conditions. European Journal of Applied Physiology, 55, 330-333.

LaPorta, M. A., Linde, H. W., Bruce, D. L. & Fitzsimons, E. J. (1978). Elevation of creatine phosphokinase in young men after recreational exercise. JAMA, 239(25), 2685-2687.

Lott, J. A. & Landesman, P. W. (1984). The enzymology of skeletal muscle disorders. CRC Crit Rev Clin Lab Sci, 20(2), 153-171.

Maxwell, J. H. & Bloor, C. M. (1981). Effects of conditioning on exertional rhabdomyolysis and serum creatine kinase after severe exercise. Enzyme, 26, 177-181.

McCellan, G., Weisberg, A. & Winegrad, S. (1983). Energy transport from mitochondria to myofibril by a creatine phosphate shuttle in cardiac cells. Am. J. Physiol. 245, c423-427.

McCully, K. K. & Faulkner, J. A. (1985). Injury to skeletal muscle fibers of mice following lengthening contractions. J. Appl. Physiol., 59(1), 119-126.

McCully, K. K. & Faulkner, J. A. (1986). Characteristics of lengthening contractions associated with injury to skeletal muscle fibers. J. Appl. Physiol., 61(1), 293-299.

Meyer, R. A., Sweeney, H. L. & Kushmerick, M. J. (1984). A simple analysis of the "phosphocreatine shuttle". Am. J. Physiol., 246, c365-377.

Nanji, A. A. (1983). Serum creatine kinase isoenzymes: A review. Muscle and Nerve, 6, 83-90.

Newham, D. J., Jones, D. A. & Edwards, R. H. T. (1983). Large delayed plasma creatine kinase changes after stepping exercise. Muscle and Nerve, 6, 380-385.

Newham, D.J., Jones, D. A., Tolfree, S. E. J. & Edwards, R. H. T. (1986). Skeletal muscle damage: A study of isotope uptake, enzyme efflux and pain after stepping. European Journal of Applied Physiology, 55,106-112.

Newham, D. J., Jones, D. A. & Edwards, R. H. T. (1986). Plasma creatine kinase changes after eccentric and concentric contractions. Muscle and Nerve, 9, 59-63.

Nicholson, G. A., Morgan, G. J., Meerkin, M., Strauss, E. R. & Mcleod, J. G. (1986). The effect of aerobic exercise on serum creatine kinase activities. Muscle and Nerve, 9, 820-824.

Nicholson, G. A., Morgan, G., Meerkin, M., Strauss, E. & Mcleod, J. G. (1985). The creatine kinase reference interval: An assessment of intra- and inter-individual variation. Journal of Neurological Sciences, 71, 225- 231.

Nicholson, G. A., McLeod, J. G., Morgan, G., Meerkin, M., Cowan, J., Bretag, A., Graham, D., Hill, G., Robertson, E. & Sheffeild, L. (1985). Variable distributions of serum creatine kinase reference values. Journal of Neourological Sciences, 71, 233-245.

Noakes, T. D., Kotzenberg, G., McArthur, P. S. & Dykman, J. (1983). Elevated serum creatine kinase MB and creatine kinase BB-isoenzyme fractions after ultra-marathon running. Eur. J. Applied. Physiol. 52, 75-79.

Norton, J. P., Clarkson, P. M., Graves, J. E., Litchfeild, P. & Kirwan, J. (1985). Serum creatine kinase activity and body composition in males and females. Human Biology, 57(4), 591-698.

Olerud, J. E., Homer, L. D. & Carroll, H. W. (1976). Incidence of acute rhabdomyolysis: Serum myoglobin and enzyme levels as indicators of muscle injury. Arch Intern Med, 136, 692-697.

Ohkuwa, T., Saito, M. & Miyamura, M. (1984). Plasma LDH and CK activities after 400 m sprinting by well-trained sprint runners. European Journal of Applied Physiology, 52, 296-299.

Ohman, E. M., Teo, K. K., Johnson, A. H., Collins, P. B., Dowsett, D. P., Ennis, J. T. & Horgan, J. H. (1984). Cardiospecific creatine kinase after strenuous exercise in female athletes. J. Sports Med. 24, 270-272.

Panteghini, M., Bonora, R., Pagani, F. & Calarco, M. (1986). Evaluation of a commercial immunoenzymemetric assay kit for creatine kinase MB isoenzyme determination using monoclonal antibodies. J. Clin. Chem. Clin Biochem, 24, 97-102.

Panteghini, M., Cuccia, C., Malchiodi, A., Calarco. M. & Pagnoni, N. (1986). Isoforms of creatine kinase MM and MB in acute myocardial infarction: a clinical evaluation. Clinica Chimica Acta, 155, 1-10.

Pedinoff, S. & Sandhu, R. S. (1978). Electromyographic effect on serum creatine phosphokinase in normal individuals. Arch Phys Med Rehabil, 59, 27-29.

Poole, D. C. & Gaesser, G. A. (1985). Response of ventilatory and lactate thresholds to continuous and interval training. Journal of Applied Physiology, 1115-1121.

Preist, J. B., Oei, T. O. & Moorehead, W. R. (1982). Exercise-induced changes in common laboratory tests. A. J. C. P., 77(3), 285-289.

Robinson, D. & Williams, P. T. (1982). Raised creatine kinase activity and presence of creatine kinase MB isoenzyme after exercise. British Medical Journal, 285, 1619-1620.

Rogers, M. A., Stull, A. & Apple, F. S. (1985). Creatine kinase isoenzyme activities in men and women following a marathon race. Medicine and Science in Sports and Exercise, 17(6), 679-682.

Ross, J. H., Attwood, E. C., Atkin G. E. & Villar, R. N. (1983). A study on the effects of severe repetitive exercise on serum myoglobin, creatine kinase, transaminases and lactate dehydrogenase. Quaterly Journal of Medicine, 206, 268-279.

Roti, S., Iori, E., Guiducci, U., Emanuele, R., Robuschi, G., Bandini, P., Gnudi, A. & Roti, E. (1981). Serum concentrations of myoglobin, creatine phosphokinase and lactic dehydrogenase after exercise in trained and untrained athletes. J. Sports Med., 21, 113-118.

Rumley, A. G., Pettigrew, A. R., Colgan, M. E., Taylor, R., Grant, S., Manzie, A., Findlay, I., Dargie, H. & Elliott, A. (1985). Serum lactate dehydrogenase and creatine kinase during marathon training. British Journal of Sports Medicine, 19(3), 152-155.

Sahlin, K., Harris R. C. & Hultman, E. (1975). Creatine kinase equilibrium and lactate content compared with muscle pH in tissue samples obtained after isometric exercise. Biochem. J., 152, 173-180.

Saks, V. A., Rosenshtraukh, L. V., Smirnov, V. N. & Chazov, E. I. (1978). Role of creatine phosphokinase in cellular function and metabolism. Can. J. Physiol. Pharmacol., 56, 691-706.

Sanders, T. M. & Bloor, C. M. (1975). Effects of repeated endurance exercise on serum enzyme activities in well-conditioned males. Medicine and Science in Sports, 7(1), 44-47.

Savabi, F., Geiger, P. J. & Bessman, S. P. (1984). Myofibrillar end of the creatine phosphate energy shuttle. Am. J. Physiol., 247, 424-432.

Schnohr, P., Grande, P. & Christiansen, C. (1980). Enzyme activities in serum after extensive exercise, with special reference to creatine kinase MB. Acta Med Scand, 208, 229-231.

Secher, N.H. (1983). The physiology of Rowing. Journal of Sports Medicine, 1 23-24.

Shumate, J. B., Brooke, M. D., Carroll, J. E. & Davis, J. E. (1979). Increased serum creatine kinase after exercise: A sex-linked phenomenon. Neurology, 29, 902-905.

Sjostrom, M. (Karger, Basel, 1982). Muscle fine structure and less advanced ischemia. Value of morphological analysis; in Lewis. pp. 80-100.

Sjostrom, M. & Friden, J. (Karger, Basel, 1984). Muscle soreness and muscle structure. Medicine and Sport Science, 17, 169-186.

Skillen, A. (1984). Clinical biochemistry of lactate dehydrogenase. Cell. Biochem. Funct., 2, 140.

Stansbie, D., Aston, J. P., Dallimore, N. S., Williams, H. M. S. & Willis, N. (1983). Effect of exercise on plasma pyruvate kinase and creatine kinase activity. Clinica Chimica Acta, 132, 127-132.

Staubli, M., Roessler, B., Kochli, H. P., Peheim, E. & Straub, P. W. (1985). Creatine kinase and creatine kinase MB in endurance runners and in patients with myocardial infarction. Eur J Appl Physiol, 54, 40-45.

Symanski, J. D., McMurray, R. G., Silverman, L. M., Smith, B. W. & Siegel, A. J. (1983). Serum creatine kinase and CK-MB isoenzyme responses to acute and prolonged swimming in trained athletes. Clinica Chimica Acta, 129, 181-187.

Tiidus, P. M. & Ianuzzo, C. D. (1982). Effects of intensity and duration of muscular exercise on delayed soreness and serum enzyme activities. Medicine and Science in Sports and Exercise, 15(6), 461-465.

Thomson, W. H. S., Sweetin, J. C. & Hamilton, I. J. D. (1975). ATP and muscle enzyme efflux after physical exertion. Clinica Chimica Acta, 59, 241-245.

APPENDIX 1

REVIEW OF LITERATURE

To fully appreciate the serum creatine kinase (CK) response to exercise a global understanding of the enzyme's structure and function is fundamental. It has been speculated that both these parameters play a prominent role in the manifestation of CK clearance witnessed post exercise. The augmented TCK activity evident following exercise has been shown to be intimately related to the delayed muscle soreness (DMS) experienced on the completion of exercise. This review will examine how factors such as: contraction type, sex, exercise intensity and duration, and, level of training effect the TCK activity witnessed post exercise. Furthermore, a review of the mechanisms involved in the TCK clearance will be examined in an attempt to elucidate the significance of the TCK response to exercise of a high intensity and short duration.

#1
CK STRUCTURE

CK exists in the cytoplasm of cells in human tissues as a dimer composed of two subunits, M and B, with a relative molecular mass of 43,000 and 44,500 respectively (Panteghini et al., 1986). These subunits combine to produce three dimeric molecular forms designated CK-MM, CK-MB and CK-BB. A fourth isoenzyme has been

isolated from the mitochondria of mammalian tissues which has been shown to differ from the cytoplasmic isoform (Nanji et al., 1983). In human sera, it is possible to resolve three CK-MM (MM1, MM2 and MM3) and two CK-MB (MB1 and MB2) subspecies (Saks et al., 1978; Bias, 1982). The CK-MM subspecies isoforms will be discussed in more detail at a later point in this review.

#2
CK FUNCTION

2.1 CREATINE PHOSPHATE ; AN IMMEDIATE ENERGY SOURCE

The performance of muscular work is dependent on the energy derived from chemical reactions within the tissues (Bessman et al., 1981).To date, there is very little information on metabolic responses of humans to brief durat[redacted]maximal exercise. During high intensity, short duration work the need for energy exceeds the energy suppled from aerobic sources, consequently, under these circumstances energy must be derived from high energy phosphagen stores (creatine phosphate) and/or anaerobic glycolysis.

In muscular work creatine phosphate (CP) is broken down to creatine and inorganic phosphate with a concomitant release of energy. This reaction is rapidly catalyzed by CK which facilitates the transfer of a high energy phosphate to ADP and thereby forming ATP. Any ADP produced from ATP hydrolysis is immediately rephosphorylated to ATP (Saks et al., 1978). CP is considered to be an immediately available store of chemical energy in cardiac and

skeletal muscle cells which maintains the intercellular ATP concentrations during periods of prolonged contraction (McCellan et al., 1983). The CP concentration in normal metabolizing cells is 3 to 4 times that of ATP. It is speculated that with intense exercise the greatest reduction in ATP occurs in type-II fibers. Research has shown that the rate of CP breakdown is dependent on the intensity of the muscular work involved. CP stores are depleted with maximum sprinting in five to seven seconds (Cheetman et al., 1986; Hirvonen et al., 1987). At the point when CP stores are depleted, energy must be derived from glycolytic means if the work is to continue. However, the energy production rate of high energy phophagen stores is much greater than that observed in glycolysis. Therefore, when CP stores are depleted to a certain level the total energy production in the form of ATP is noticeable reduced. The critical level of CP is reached when CK is no longer saturated with the substrate CP (Hirvonen et al., 1987).

Maximal cycling exercise of a brief duration results in a marked decrease in muscle phosphagens and glycogen with a concomitant increase in glycolytic intermediates (Hirvonen et al., 1987). In performing a thirty second sprint, marked decreases in glycogen are recorded. Nevertheless, with exercise of increased intensity and shorter duration, subjects become exhausted before glycogen depletion occurs (Cheetman et al., 1986).

2.2 CK REACTION KINETICS

CK, also referred to as creatine phosphokinase, catalyzes the reversible phosphorylation of creatine by adenosine triphosphate (ATP). The optimal pH values for the forward (CR + ATP --> ADP + CP) and reverse (CP+ ADP --> ATP + CR) reaction are 9.0 and 6.8, respectively. The equilibrium position for the reaction is pH dependent. At neutral pH, CP has a much higher phosphorylation potential than does ATP; this favors the reverse reaction, with ATP being formed from CP (Bias et al., 1982). The apparent equilibrium constant at pH 7 is as follows:

$$k' = \frac{[ATP]\ [creatine]}{[ADP]\ [CP]} = 100$$

In a sense the reaction illustrated above is a dead end reaction , as there is no other known reaction involving CP in the cell (Sahlin et al., 1975). The aforementioned reaction is very near equilibrium in skeletal muscle (Bessman et al., 1981). The equilibrium constant and ATP:ADP ratio are such that when high energy hydrolysis occurs, the CP pool is depleted to a greater extent than ATP (Saks et al., 1978).

The km for the ATP + CR side of the reaction equation is much higher than that of the ADP + CP side. Although the reverse reaction is kinetically preferable, oxidative phosphorylation accelerates the forward reaction in the mitochondria (McCellan et al., 1983 , Meyer et al., 1984). CP is phosphorylated in the mitochondria directly from ATP generated by oxidative phosphorylation.

The equilibrium constant for myofibrillar CK is very low, as a result, ATP production from ADP and CP is kinetically preferable (Saks et al., 1978). In this situation the equilibrium of the reaction is such that it acts to transfer a phosphate from CP to ADP yielding ATP at the CK myofibril site on the M-line (Houk & Putman,1973 & Savabi et al.,1984).

2.3 THE ATPASE /CK INTERACTION

The molecular mechanism of muscular contraction involves ATP hydrolysis in the actomyosin ATPase reaction (Saks et al., 1978). Available kinetic and biochemical evidence have indicated a possible functional interaction between myofibril CK and ATPase, whereby the product of the ATPase reaction is utilized as substrate for the CK reaction (Savabi et al., 1984). In the presence of CR, the ADP produced from ATP hydrolysis during muscle contraction is immediately rephosphorylated by the myofibrillar CK reaction at the expense of CP . The newly produced ATP is then used in the subsequent contraction cycle (Meyer et al., 1984). Extensive muscular work requires an effective energy supply and rapid removal of the immediate product of the ATPase reaction, (ADP), to avoid its accumulation and the inhibition of the ATPase (Meyer et al., 1984; McCellan et al.,1983). ATPase and CK are thought to be spatially coupled in such a way that energy rich phosphates are efficiently transferred by CK to the ADP formed by ATPase. Research has demonstrated a high ATP:ADP ratio is found in the vicinity of the ATPase (Houk & Putman, 1973). Although, the high ATP

concentrations in the myoplasm are not easily accessible for ATPase. The newly formed ATP is compartmentalized in such a manner that it gains preferential access to the active site (Bessman et al., 1980). This close functional relationship between CK and ATPase could possibly be due to a physical association between these two enzymes, which serves an important role in energy production and utilization (Erickson-Viitanen et al., 1982).

2.4 CK LOCATIONS

CK has been shown to bind to the outer side of the mitochondrial matrix, myofibril, sarcoplasmic reticulum (S.R.) and sarcolemma. However, the two most prominent sites of its attachment are the myofibril and mitochondria (Bessman et al., 1981). CK bound to the mitochondria accounts for 40% of the total CK activity of the cell (Saks et al, 1978). The myosin head on the M-line has been demonstrated to contain the major CK-MM isoenzyme component (Bessman & Fonyo, 1966). On contraction the myosin and actin heads are brought into a region rich in CP located around the I-Band. This provides substrate for the transfer of phosphate to the bound ADP at the crossbridge site and thus permits relaxation to occur (Savabi et al., 1984). The CK that is associated with the S.R. is in close proximity with the Ca^{2+} dependent ATPase. The ATP utilized by the Ca^{2+} pump is used for Ca^{2+} sequestering during active relaxation in skeletal and cardiac muscle (Saks et al., 1978). The CK located on the plasma membrane play's a similar role to that of sarcoplasmic reticulum CK, in that, the ATP produced in the CK

reaction is utilized in supporting ion balances across the surface membrane of the cell (Bessman et al.,1981). It has been suggested that energy transfer from the mitochondria to target organelles such as the sarcolemma, S.R., and myofibrils is organized by a CP energy shuttle (Meyer et al.,1984). The two prominent locations of CK provide appropriate receptors for the CP shuttle at both the mitochondrial and myofibrillar ends.

2.8 THE ROLE OF THE CP SHUTTLE

CP is thought to play an important role as a buffer for the resynthesis of ATP in muscle metabolism. CK isoenzymes are instrumental in the intercellular energy transport from the mitochondria to the myofibrils and other sites of energy utilization (Houk & Putman et al., 1973). Large cellular pools of ATP may not be directly accessible for utilization in muscle contraction. Instead only a small pool of ATP around or in the myofibril may be utilized. These small pools of ATP are replenished by CP which has been suggested to be a high energy phosphate carrier connecting the mitochondrial pools of ATP with the myofibrillar and possibly other ATP pools (Saks et al., 1978). The M-Band is believed to use the CP that has diffused along the I-Band from the mitochondria (Bessman et al., 1981). The apparent preferential utilization of CP by myofibrillar ATPase supports the idea that energy for muscular contraction is delivered through a mitochondrial - myofibrillar shuttle of CP and CR. For the shuttle to operate there must be CK present in both the mitochondria and the myofibril (McCellan et al.,

1983). The mitochondria synthesizes ATP from ADP and Pi. Creatine is phosphorylated in the mitochondria to CP by means of the CK reaction. CP diffuses to the myoplasm where a second CK pool rephosphorylates bound ADP to ATP transferring a phosphate from CP (Bessman & Geiger, 1981).

Thus, CP has classically been considered to function as a storage form of high energy phosphates that buffer changes in ATP and ADP levels, however, it has is been proposed that CP and CR function as a "shuttles" for the transport of high energy phosphates between compartments of adenylates within the muscle cell.

2.6 CK INTERPLAY WITH OXIDATIVE AND GLYCOLYTIC METABOLISM

CP acts as an energy donor for contraction in which glycolysis and oxidative phosphorylation supply the energy for its rephosphorylation. The action of CK produces the immediate product of ADP which acts to stimulate oxidative phosphorylation to produce CP (Savabi et al., 1984). Some believe that the CK reaction is a side reaction in the supply of energy for contraction, whereby ATP from glycolysis and oxidative phosphorylation is considered to be used directly in the myofibrillar ATPase reaction, while CP acts purely as a high energy phosphate store (Saks et al., 1978; Meyer et al.,1984).

#3
CK RESPONSE TO PHYSICAL ACTIVITY

Tiidus et al.,(1983); Galteau et al., (1976) & Berg (1978) report that the magnitude of the serum TCK response is directly proportional to the intensity and duration with which an activity is performed. Although, the intensity of the exercise was found to have a more pronounced effect on the enzyme response than the duration. Lott & Landesman, (1984); Fowler et al., (1968); Chahine et al., (1976) and Hunter & Critz 1971 refute this finding. They contend that increases in CK activity are linearly related to the duration of the exercise interval.

Females have been shown to have lower resting CK levels than males. Rogers and co-workers research in 1985 demonstrated that females had a significantly lower serum CK activity than the males at all points of sampling post marathon. Shumate et al., (1979) contends that female skeletal muscle is less sensitive to exercise than males. Norton et al., (1985) and Rogers et al.(1984) suggests that this may be attributed to differences in lean body mass seen between males and females. Since skeletal muscle contains the highest activity of CK in the body, (CK-MM 90-100%, CK-MB 0-10%) the observed male-female sex differences in serum CK elevation could be attributed to a larger muscle mass in men. In addition, higher CK activities in males may reflect a larger muscle fiber recruitment which in turn facilitates greater skeletal muscle leakage of CK into the circulation (Rogers et al., 1985; Apple et al.,

1986). Kirwan et al.,(1986) contends that decreases in the CK response after exercise in females could be associated with an estrogen mechanism which safe guards females from exercise-induced muscle damage. This increased levels of estrogen in female may reduce enzyme effluxing due to the protective or stabilizing effect it has on the sarcolemma (Norton et al., 1985).

3.1 CK RESPONSE TO AEROBIC TRAINING

Elevated serum enzyme activities in healthy well trained long distance runners may suggest that prolonged strenuous exercise results in substantial damage to skeletal muscle (Rogers et al., 1985). Recent documentation of changes in the ultra structure of skeletal muscle from marathon runners would lend considerable support to this conclusion (Evans et al 1986; Apple et al., 1987; Hikida et al., 1983). Several reports would indicated that serum CK release after a standard load of exercise may be lessoned following a period of aerobic training (Rogers et al., 1985; Olerud et al.,1976, Brown 1983). Nevertheless, it has been noted that the trained individual displays an elevated resting TCK. Roti et al., (1981) and Ross et al., (1985) suggest that training diminishes the release of enzymes into the serum following exercise, yet augments the CK activity at rest. Both researchers contend that training elicits a cellular adaptation in the exercised tissue that facilitates an increase in muscle enzyme synthesis followed by enhanced plasma effluxing. Moreover, research shows that this effect may simply reflect relative muscle mass or increased turnover of skeletal

proteins on a long range basis in the trained individual (Priest et al., 1982; Stansbie et al., 1983). Jansson et al., (1985) reported that CK activity was not related to training status, although it was lower in type I(slow) fibers than type II(fast). It was demonstrated that the CK-MB content in skeletal muscle increased with the degree of endurance training. Schnohr and co-workers (1980) contend that there is no difference in post exercise CK levels in endurance trained versus untrained individuals.

Apple et al., (1984) has demonstrated that slow twitch fibers contain a higher percent of MB than type II fibers. Endurance training appears to increases the percentage of CK-MB in type I fibers which acts to yield a more heart-like muscle fiber. The human heart is comprised of fifteen to twenty percent CK-MB and seventy-five to eighty percent CK-MM (Apple et al., 1984). CK-MB has been correlated with the oxidative capacity of a cell as estimated by mitochondrial CK (Jansson et al., 1985). As mentioned previously, TCK activity has been shown to be higher in type II fibers. This is consistent with the suggestion that TCK action is implicated in the intercellular energy storage that should theoretically be more developed in type II fibers due to their ability to react to sudden energy demands (Kettunen et al., 1982). CK-MB elevation above six percent of the TCK is widely accepted as an indicator of acute AMI (Stansbie et al., 1983). Kettunen et al.,(1982); Lott & Landesman, (1984) and Apple et al., (1986) have reported CK-MB activities greater than six percent of the TCK in athletes post marathon. However, on further scintigraphic analysis, no damage to the myocardium was viewed. Schnohr et al. (1980) concluded that, since

trace amounts of CK-MB are found in skeletal muscle and the molecular weight of CK-MB and TCK are virtually identical, it is likely that the small amounts of CK-MB found in the serum post-exercise originate from skeletal muscle sources. In fetuses and in dystrophy patients developing and regenerating muscles contain a higher CK-MB content than muscles of sedentary individuals (Lott & Landesman, 1984). Researchers believe that the damaged skeletal muscle begins its regeneration process through a reversal back to its fetal form (Apple et al., 1987). Stress induced by training might reactivate the 'B' subunit gene to initiate and increase the synthesis of MB, BB or both (Apple et al., 1984). Rogers et al., (1985) suggests that chronic runners have a transient rhabdomyolysis which during recovery leads to muscle regeneration through the fetal state. Chronic stress-induced injury to skeletal muscle may allow for CK isoenzyme composition to differentiate through a pattern similar to embryonic skeletal muscle formation by degeneration and regeneration of CK BB to MB to MM. This chronic state of fiber necrosis and regeneration could lead to an increase in CK -MB content in the skeletal muscle (Apple et al., 1987). It is possible that the CK-MB isoenzyme is released selectively from different fiber types within the injured muscle (Newham et al., 1986). Moreover, injury to the enriched CK-MB muscle would augment the levels of the heart like isoenzymes cleared into the circulation and thereby yield an enzymatic profile similar to those observed following AMI.

3.2 DELAYED MUSCLE SORENESS (DMS) AND THE CK RESPONSE

Heavy physical work performed by trained or untrained individuals results in delayed muscular pain and stiffness. This delayed muscle soreness (DMS) has been attributed to the eccentric component of foreign exercise (Kasperek & Snider, 1985). To date there is no general agreement on the pathophysiological mechanism responsible for DMS. Delayed muscle soreness is a concept with a variety of meanings. In brief, it comprises a number of symptoms with varying character and degrees of difficulties (Friden et al., 1986). The underlying pathophysiological processes which are usually fiber-type specific are very different and depend on the amount and type of work carried out (Sjostrom & Friden, 1984).

Both CK and myoglobin are used as objective indicators of muscle trauma (Olerud et al., 1976). Myoglobin appears earlier in the circulation than CK following exercise due to its smaller molecular weight (17,800 as compared to 80,000) (Byrnes et al., 1985). Myoglobinanemia (detection of myoglobin in the serum) is thought to be the most sensitive indicator of micro-trauma to skeletal muscle (Maxwell et al., 1981; Olerud et al., 1976; Demos et al., 1974). However, CK which is predominantly composed of the skeletal muscle type isoenzyme (MM) has been demonstrated to be an accurate enzymatic marker of skeletal muscle trauma (Lott & Landesman,1984 ; Ross et al., 1987).

Following heavy physical work of an eccentric nature muscles exhibit Z-band disorganization, I-band widening and disruption of

the sarcomeres (Newham et al .,1986) High myofibrillar tension development during contraction causes a mechanical disruption of the Z-disc (Friden , 1984). During the period of overload, the Z-disc has been shown to be the weak link in the myofibrillar contractile chain (Friden et al., 1986). Elevated lysosomal enzyme activity and calcium (Ca^{2+}) levels have been reported to weaken this particular portion of the myofibril structure (Armstrong, 1984). Lysosomal enzymes are implicated in the increased rate of muscle protein degradation observed with exercise. This increase in lysosomal activity is likely due to macrophage infiltration associated with degenerative (proteolysis) processes occurring in damaged muscle (Kasperek & Snider, 1985; Nicholson et al., 1986; Abraham ,1977).

As mentioned previously eccentric exercise has been shown to damage the sarcolemmic architecture. It has been well documented that eccentric work can lead to a very large loss of muscle soluble enzymes (notably CK) into the circulation (Armstrong et al., 1984). While most exercise consists of eccentric and concentric contractions, it is believed that the eccentric component is chiefly responsible for causing this muscle damage (Evans et al., 1986). The difference in the extent of tissue injury witnessed post exercise may be attributed to either contraction strength and/or velocity (McCully & Faulkner, 1985). Thus, muscle pain and tenderness develops preferentially after eccentric work.

3.3 MECHANISM OF CK CLEARANCE AND ALTERATIONS

The mechanism of enzyme clearance, which is yet unknown may include removal of enzymes by the reticuloendothelial system, inactivation by the lymphatic system, clearance of enzymes by the tissue of origin and partial degeneration by the enzyme in the cell (Rogers et al., 1985). In view of known differences in the muscular contraction types, the mechanism responsible for the enhanced TCK clearance witnessed post exercise is controversial. DMS and enhanced CK clearance has been attributed to: (a) metabolic effect on muscle fibers producing increased membrane permeability and/or (b) mechanical effect on muscle fibers resulting in membrane damage and fiber necrosis (Newham et al., 1986). Whole body massage which employs mechanical manipulation of the muscle has been shown to elicit increases CK activity, however the mechanism is uncertain (Clarkson et al., 1985).

The CK efflux demonstrated with exercise may be due to metabolic factors, however concentric contractions are known to be metabolically more costly than eccentric contractions (Newham et al., 1983). Moreover, eccentric work produces greater tension per active motor unit than corresponding concentric work (Friden et al., 1986), which in turn is speculated to evoke a profound mechanical disruption to the exercising tissues (Sjostrom & Snider, 1984). Activities such as downhill running and eccentric stepping have been found to induce DMS due to their bias to eccentric contraction. (Clarkson et al., 1985; Byrnes et al., 1985; Newham et al., 1983). Symanski et al., (1983) observed no significant increase in TCK

activity with trained swimmers following acute and prolonged swim stress testing to the point of exhaustion. These findings could perhaps be attributed to a decrease in mechanical force produced in swimming as compared to running which would suggest that under these circumstances CK release is related to mechanical factors rather than metabolic fatigue .

The release of CK from the muscle has been shown to occur relatively suddenly and only after prolonged periods of exercise characterized by marked decrease in intercellular ATP levels (Schmit & Schmit, 1968). If the exercise is not sufficient in depleting the intercellular levels of ATP, one does not see these pronounced increase in enzyme fluxes across the sarcolemma (King et al., 1976). An initial decline in ATP stores occurs at the onset of exercise, although, only prolonged and strenuous exercise will depress the ATP levels low enough to allow for the effluxing of the intercellular enzymes (Thomson et al., 1975). Olerud and co-workers (1976) speculated that during extreme exercise, energy deficiencies occur within the cell that may result in subcellular competition for the energy-rich phosphates. Such competition could result in a relative deficiency in structural maintaining energy. Maxwell et al., (1981) suggests that serum elevations of muscle enzymes following strenuous exercise is due to leakage through damaged cell membrane compromised by a depletion of ATP that is normally dedicated to the maintenance sarcolemmic integrity .

Hunter and Critz (1971) reported that endurance training reduces the CK response to maximal and submaximal exercise loads. It was suggested that trained muscle can produce ATP more

efficiently than untrained. Therefore, the release of enzymes post exercise occurs later and only after greater degree of fatigue in the trained individual (Priest et al., 1982). Trained muscle has also been shown to have an increased availability of ATP, which may help in maintaining the integrity of the cell membrane during strenuous work (Stansbie et al., 1985). Holloszy (1971) noted that endurance training augments the synthesis of particular enzymes and increases the size and number of mitochondria. Since the mitochondria supplies approximately ninety-five percent of the skeletal muscle ATP by oxidative phosphorylation, the capacity of skeletal muscle for producing ATP should be enhanced (Cheetman et al., 1986). The increased amount of ATP available during strenuous exercise may assist in maintaining the integrity of the cell membrane, thereby rendering the cell wall more stable and thus reduce the enzymatic fluxing observed post exercise.

When total work remains relatively constant, serum enzyme levels reflect cellular damage and repair (Newham et al., 1983). An intriguing and as yet unexplained feature of DMS is that the maximal enzyme release occurs usually some time after the exercise was performed, a time course that is appreciably longer than that of muscle pain (Newham et al., 1986). Evans et al.,(1986) noted that myofibrillar damage is greater three days after eccentric exercise than immediately following. This would indicate that the degradation of the contractile unit is a delayed event. Newham et al., (1983) found that muscle tenderness was decreasing at a time when CK activity was reaching its peak. This suggests that a pain producing substance might act as a precursor for the subsequent rise

in enzyme release. The processes associated with tissue repair could be factors contributing to the soreness sensation viewed post exercise. The following chain of events is speculated be involved in the DMS response.

a. High tension eccentric contractions lead to structural damage

b. Cell membrane damage leads to disruption of the calcium ion homeostasis and enzyme leakage.

c. Products of macrophage activity and intercellular contents accumulate in the interstitium, which in turn stimulates free nerve endings in the muscle leading to the sensation of DMS.

(Armstrong, 1984; Tiddus et al., 1983; Bloor, 1978)

Armstrong et al., (1984) suggests that training for a specific activity reduces the DMS response. The etiology and cellular mechanisms responsible for this reduction are unknown. There appears to be a great deal of variation between subjects sensitivity to eccentric work. Training appears to be highly specific, not only for the particular muscles involved but also for the predominant type of contraction employed in the event. Some individuals are habitually eccentrically trained. Thus, the extent to which the active muscle is lengthened and the degree of habituation to eccentric work can play an prominent role in determining the nature of the enzyme response (Newham et al., 1986).

#4
ADVANCED MEANS BY WHICH TCK ALTERATION CAN BE CHARACTERIZED

The release of muscle enzymes has been a valuable means of assessing the extent and time course of muscle damage, yet, because it is measured in the general circulation, it cannot give precise information about the location of the damage. A recently developed method of identifying muscle damage is to trace the uptake of radio labelled ^{99m}technetium pyrophosphate (Tc-PYP) (Newham et al., 1983). Tc-PYP uptake is used to identify those muscles that are effluxing enzymes, although, little is known about the mechanism by which Tc-PYP is taken up by the damaged tissue. One possible explanation is via hyperelemia and altered capillary permeability (Newham et al., 1986). Perhaps the radio isotope diffuses through non-specific lesions in the muscle fiber membrane at the same time that a soluble enzyme leaks out (Ohman et al., 1984). Thus, Tc-PYP tracings can provide useful information about the localization and time course for muscle damage. However, it is possible that a form of muscle damage occurs in which no enzyme are released from the muscle (Newham et al., 1985).

As was mentioned previously CK-MM exists in three isoenzyme forms in the plasma. CK-MM 1 is the pure gene product and is released from from the muscle tissue into the circulation. Once in the circulation, the MM 1 is converted into the isoform MM 2 and then MM 3 (Clarkson et al., 1985). Elevated levels of the MM 1 isoform in the blood indicates new release of CK into the plasma prior to its conversion. The MM 1/ MM 3 ratio can provide an index of enzyme

release rate from damaged tissue. Clarkson et al., (1987) states that the conversion of CK-MM1 to MM2 to MM3 is is performed rapidly in the blood, as it is in the MM3 form that CK must be cleared from circulation. The presence of small fractions of CK-MM1 at rest reflects release of CK from muscle tissue undergoing normal physiological turnover (Clarkson et al., 1987). An increase level of MM1 in the circulation can provide a sensitive indicator of enzyme release from the exercised muscle. As the release of MM1 into the circulation begins to exceed the rate of clearance from the blood the total Ck activity increases markedly (Clarkson et al., 1987). Thus, the quantification of CK isoforms can provide an earlier index of CK release from damaged muscle tissue and moreover, may be useful in tracing the time course of muscle damage and repair.

#5
SUMMARY OF FACTORS THAT INFLUENCE TCK EXPRESSION

The release of CK following exercise is thought to be analogous to the release of cardiac specific enzymes after acute myocardial infarcts (AMI), As the magnitude of the enzyme flux is related to the severity of the infarct, the release of the skeletal muscle enzymes following exercise has been postulated to be related to a number of different contributing factors such as: the physical condition of the subject (Apple et al., 1987; Jansson et al., 1985; Brown, 1983; Sanders & Bloor, 1975); magnitude of the stressor (exercise intensity and duration) (Tiddus et al., 1983; Rogers et al., 1985);

subject sex (Kirwann et al., 1986; Shumate et al.,1979), specific character of the enzyme (Priest et al., 1982); lean body mass (Norton et al., 1985); choice of sampling time (Lott & Landesman,1984); assaying method utilized for CK analysis (Nicholson et al.,1985; Bias et al., 1982); and, the nature of the exercise (contraction type and mode of exercise) (Newham et al., 1986 ; Roti et al., 1981; Friden et al., 1986; Kasperek & Snider, 1985). These factors have all been demonstrated to elicit a powerful influence on the elevated enzyme clearance witnessed on completion of exercise.

It is quite apparent that the effect of exercise on certain biochemical tests can lead to false diagnosis, especially where exercise-induced muscle trauma mimics the effect of muscular disease (Nicholson et al., 1986). Since CK levels are frequently used to diagnose pathological conditions, it is particularly critical that the subjects level of physical activity be taken into account under these circumstance to avoid false diagnosis.