Original article

Reference values for creatine kinase activity in a group of high-performance athletes

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Summary

Physical activity integrates processes that are manifested in biochemical adaptations, such as the increase in the serum activity of the enzyme creatinquinasa (CK). People who participate in daily training have higher CK activity values, suggesting that those of an athlete cannot be compared with those established for healthy non athletic subjects. This study proposes possible reference intervals for male and female athletes, evaluated critical values, examine the dependence of gender on these results, and compare them with those established for healthy non athletic individuals. For this, 436 results of serum CK activity obtained from male and female athletes (age: 18-40 years) were analyzed.
Results: the medians of the mean athletes (325 U/L) present a high result (*P* <0.0001) compared to the median of the female athletes (156 U/L). 59% of the results obtained in men and 39 % in women exceed the reference value established for healthy subjects. (men: 32-294 U/L; women: 33-211 U/L). The 2.5% and 97.5% percentiles and their 90% confidence intervals were calculated (men: 88 (56-90) to 833 (781-973) U/L; women: 58 (44-63) to 448 (433-497) U/L). Comparing the results with the reference values used by the laboratory for healthy subjects, significant differences were observed, with higher values obtained for the groups of athletes. From the analysis carried out, specific reference intervals were obtained, whose limits are higher than those already established, and differ by sex, being higher in men than in women. The value of biochemical expertise in the health control within their training plan is relevant to organize the distribution of workloads, prevent injuries and ensure health care.

Key words:
Adaptation. Enzymatic activity. Creatine kinase. Reference intervals.

Valores de referencia de actividad de creatinquinasa en un grupo de deportistas de alto rendimiento

Resumen

La actividad física integra procesos que se manifiestan en adaptaciones bioquímicas, como el incremento de la actividad sérica de la enzima creatinquinasa (CK). Quienes participan de entrenamientos diarios poseen valores de actividad de CK elevados, sugiriendo que los de un deportista no pueden compararse con los establecidos para sujetos sanos no atletas. Este trabajo propone intervalos de referencia posibles para atletas hombres y mujeres, evaluar valores críticos, examinar la dependencia del sexo en esos resultados, y compararlos con los establecidos para los individuos sanos no deportistas. Para ello se analizaron 436 resultados de actividad sérica de CK obtenidos de hombres y mujeres deportistas (edad: 18-40 años).
Resultados: La mediana de los deportistas varones (325 U/L) presenta un resultado mayor (p <0,0001) respecto a la mediana de las deportistas mujeres (156 U/L). El 59% de los resultados obtenidos en varones y el 38% en mujeres supera el valor de referencia establecido para sujetos sanos (varones: 32-294 U/L; mujeres: 33-211 U/L). Se calcularon los percentiles 2.5% y 97.5%, y sus intervalos de confianza 90% (varones: 88 (56-90) a 833 (781-973) U/L; mujeres: 58 (44-63) a 448 (433-497) U/L). Comparando los resultados con los valores de referencia utilizados por el laboratorio para sujetos sanos, se observaron diferencias significativas, con valores más altos para los grupos de deportistas. Del análisis realizado se obtuvieron intervalos de referencia específicos para esta población, cuyos límites son superiores a los ya establecidos, y difieren por sexo, siendo más altos en varones que en mujeres. El valor de la experticia bioquímica, en el control de salud a deportistas dentro de su plan de entrenamiento resulta relevante para organizar la distribución de cargas de trabajo, prevenir lesiones y asegurar el cuidado de su salud.

Palabras clave:
Adaptación. Actividad enzimática. Creatinquinasa. Intervalos de referencia.

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Introduction

Physical activity involves a series of mechanical, psychological, emotional and physiological processes that are expressed in biochemical changes and adaptations in order to get sufficient energy to perform the activity. One of these processes is the increase in serum activity of the creatine kinase enzyme (CK, EC 2.7.3.2.), which is released into the blood stream from muscular fibres, due to repeated intense contractions.

In these conditions, CK activity increases considerably, and although its validity as a biochemical marker of a real muscular lesion induced by exercise is still questioned by some authors, its use as such is accepted by most. Its highest serum values are attained between 12 and 24 hours after exercise and remain high for up to 96 hours or more, depending on the stopping of the stimulus[1-5].

Individuals taking part in training programmes have higher CK activity values at rest than people who are not in a programme, although this response is individual and depends on the training model and the type of contraction performed. Furthermore, this condition is alleviated by adapting the repetition of exercises, which lowers the quantity and quality of muscular lesions. Determining the serum CK activity levels among athletes is a very useful biochemical marker to estimate the stress caused by the training on the muscle, suspect cases of over-training, prevent lesions from appearing and monitor the post-effort muscular recovery processes. In apparently healthy subjects, CK levels which exceed the reference value might be correlated with their physical condition; however, if these levels persist at rest, this might be a sign of subclinical muscular disease, demonstrated through the appearance of symptoms such as chronic fatigue[4,6-9].

Within this situation, we should mention the need to get reference values from the actual groups of athletes. The reference value concept was proposed by the IFCC panel of experts and accepted with different nuances by the national commissions for the purposes of unifying concepts, methods, terminologies and conducts. A reference value is defined as a value obtained from a reference individual, a person within the community, who has a determined state of health. All individuals who meet the defined inclusion conditions constitute a reference population which makes it possible to set values for specific groups[10-12].

It is intriguing to think that CK activity results for an individual doing their sport cannot be compared with the reference intervals determined for healthy non-athlete subjects. We might therefore ask which value would be reasonable to expect as a physiological response in athletes without considering an associated pathology. The answer to this question has great practical value, as if a CK result has increased over the reference value in an athlete within what is stipulated as an appropriate response, the training load might even be increased without risking an additional lesion, in search of greater physical and physiological responses. On the contrary, if the value was very high, it would be advisable to reduce loads to protect the athlete and prevent possible muscular lesions.

The goals of this paper were to propose possible reference intervals in male and female athletes between 12 and 24 hours after training, examine how much these results depend on gender, compare them to the results determined for healthy non-athletes and assess critical values that should raise the alarm among treating physicians.

Material and method

A cross-sectional, descriptive and retrospective design was produced which analysed 436 results obtained over a period of 10 years (March 2009-November 2019), from male and female athletes aged between 18 and 40 years old, which were sent to the LACBA S.A. Laboratory at the TCBA Diagnostic Centre for routine biochemical control They were all high-performance hockey players who went to training sessions lasting 60-120 minutes, six days a week with one rest day per week. On admission, they were asked to fill in a form asking if they considered that they met the criteria to be included in this study, alongside the form asking for informed consent to use the results for scientific purposes Inclusion criteria were the absence of previous diseases, muscular lesions diagnosed by the sports medical team, admission to hospital in the last three months or for periods over 15 days during the year prior to the sample-taking, and use of medication.

Obtaining results

The results analysed were obtained from serum samples using elbow fold venepuncture on the athletes 8-10 hours after 12 hours of fasting and sleep, with two days of prior training. The athletes abstained from training in the morning, to avoid changes in the plasma volume induced by exercise which might affect measurements. The blood sample was left to coagulate for 30 minutes at room temperature, it was centrifuged for 12 minutes at 3,500 rpm and then processed. CK serum activity was determined using the IFCC UV method in an Advia 1800 Siemens auto-analyser, and the results were expressed in U/L at 37°C. The reference values used were provided by the manufacturer (men: 32-294 U/L; women: 33-211 U/L) and verified by the laboratory. Institutional endorsement was provided to use the results obtained through the LACBA laboratory computer system, copied onto password-protected Excel sheets, saved on two pendrives which could only be accessed by the researchers.

Statistical analysis

The CK variable distributions in the groups being analysed were significantly different ($p < 0.05$ for the Kolmogórov-Smirnov test)[10,13]. Medians were compared using the Mann Whitney test between the male and female groups; and between the reference values obtained and those used by our laboratory (2.5 and 97.5 percentiles and 90% confidence interval) with the Wilcoxon test. In all cases, a <0.05 level of significance was considered. The Rout method was used to define outliers. The statistics programs used were SPSS 25.0 and GraphPad 8.

Results

The CK activity distribution in the athletes being studied, differentiated by gender (male and female), presented non-parametric behaviour, as shown in the histograms in Figures 1 and 2.

The outlier values were determined, and the descriptive statistics and 2.5% and 97.5% percentiles were calculated, and then used to calculate the respective reference values with their confidence intervals (CI 90%) (Table 1). The reference values for healthy subjects used by the laboratory were provided by the equipment and reagent manufacturer.

Figure 1. Distribution of the CK serum activity levels in male athletes.

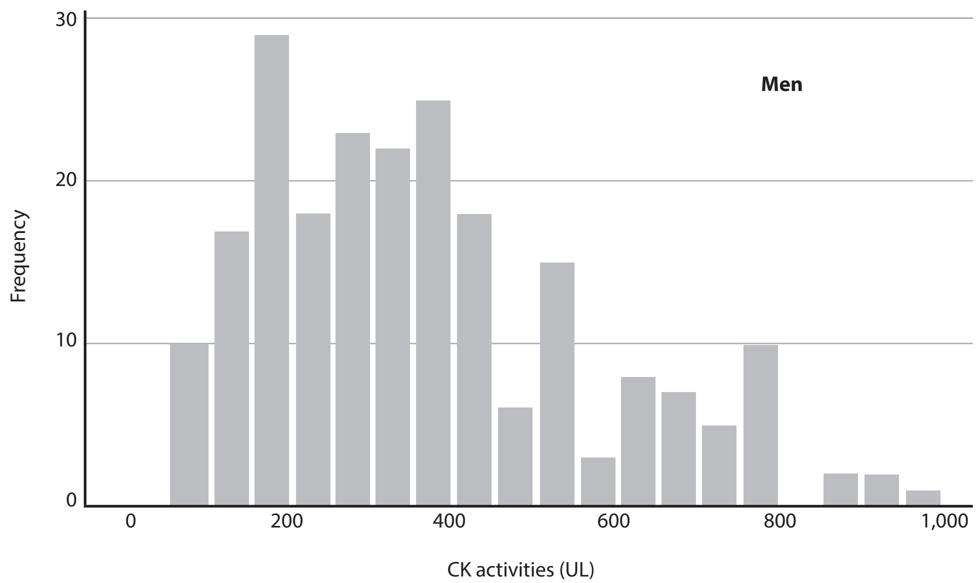
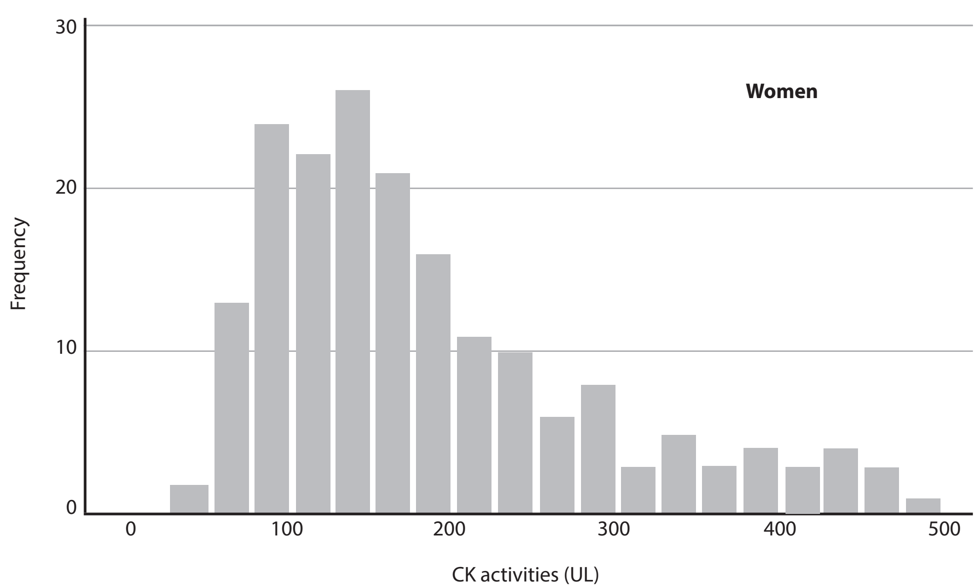


Figure 2. Distribution of the CK serum activity levels in female athletes.



The transferability was verified for these reference values from a sample population of 160 individuals (84 men and 76 women), considering that 95% of these values fall within the manufacturer's reference limits[14-16]. The comparison of the laboratory reference values with those obtained from the group of athletes was significantly different (p<0.001). It was observed that 59% of CK results obtained among men and 38% in women exceed the reference value set for healthy subjects. From the analysis of the gender-differentiated CK activity results, a statistically significant difference (p<0.0001) was observed when comparing the medians. The median of male athletes presents a higher result (325 U/L) than the median of female athletes (156 U/L).

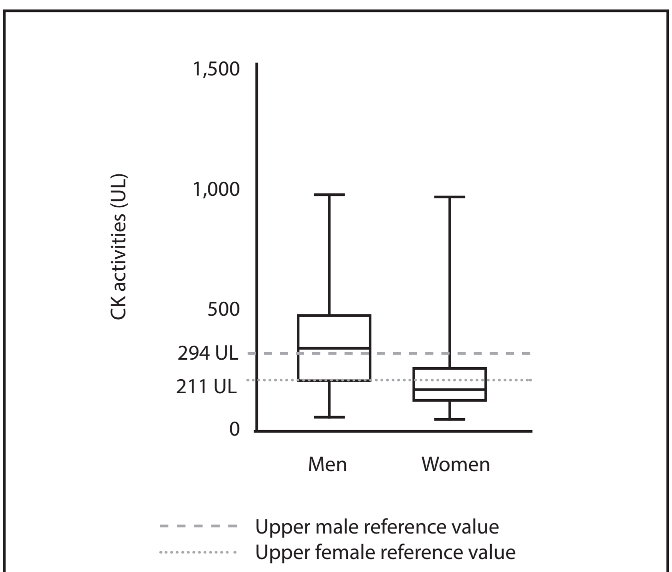
Figure 3 shows the comparison of the CK activity results by gender, detailing the boundaries of the upper reference value used by the laboratory.

Table 1. Medians and lower and upper reference values with their respective CI 90% for male and female athletes.

	Men	Women
Median U/L	325	156
Reference intervals		
Lower limit U/L	88 (CI 90%: 56-90) *	58 (CI 90%: 44-63)*
Upper limit U/L	833 (CI 90%: 781-973) *	448 (CI 90%: 433-497)*

CI 90%: confidence interval 90%
* p < 0.001.

Figure 3. Comparison of CK activity values by gender. The diagram shows the upper limits of the reference value used in the laboratory with broken lines.



Discussion

CK serum activity is an important indicator of the extent of muscular effort and adaptation to the training. It has become an exceedingly interesting option to assess athletes and a very useful tool for the medical team looking after them, although it is not easy to interpret.

This paper demonstrates that CK activity values are higher in athletes, both men and women, which can help to explain the meaning of this data by providing specific reference values for a group of athletes that can be used in sports medicine during daily practice[17,18].

It should be considered that the values obtained depend on the type of training (intensity, duration, type of muscular contraction performed), so different groups of athletes might refer to slightly different results. This study assessed male and female high-performance grass hockey players. This sport has undergone radical changes in its technical, tactical and physiological requirements, above all in elite athletes, particularly associated with the playing surfaces and the greater intensity of the game. There are few publications in the literature on muscular damage among elite grass-hockey players[19].

Given that CK activity remains high a few days after the stimulus and that the athletes studied carry out successive training sessions, the results obtained reflect the accumulative effect of the loads and the degree of adaptation to the training. In this particular case, the hockey players carry out high-intensity intermittent activities and these results might suggest that they could be extrapolated to sports with similar characteristics (such as football)[20-22].

However, we suggest that it is reasonable to assess specific reference intervals per sport. The athletes' reference limits were 2 to 3 times higher than those used by the laboratory for healthy subjects. It should be emphasised that the values generally used in the laboratory concur with the bibliography in general and with the data reported by the reagent suppliers for healthy subjects who do not do any physical activity, and that require physical activity to be stopped for 72 hours prior to the extraction as a preanalytical rigour condition.

The CK activity reference intervals in athletes demonstrated differences by gender, greater in men than in women, in concordance with what was reported by the studied bibliography and in line with the existence of specific reference values per gender in the general population[23,24]. The upper limit of the reference value obtained for the group of men demonstrated higher results than for the group of women, 1.8 times higher. These differences can be explained by different reasons, such as higher CK content in men's muscles, their greater muscle mass, the protective effect of oestrogen in women, and muscle factors such as stabilisation in the permeability of the membrane due to the hormonal influence, the rate of CK elimination and the lymphatic activity[25,26].

When comparing both groups against the reference values used for healthy subjects by the laboratory, significant differences were seen, with higher values for the groups of athletes whose upper reference limits were 2.8 times higher for men and 2.1 times for women.

There are many factors that determine the increase in enzyme activity during and after exercise. The greatest values are obtained after exercises with predominantly eccentric contractions, after very lengthy or high intensity exercises[27].

The medical team must be aware of borderline situations where these increases might be associated with the possibility of muscular lesions or with pathological situations. Beyond the reference values, the concept of alarm values then appears, understanding this to be results which must be immediately reported to the doctor so that corresponding actions can be taken to keep the athlete healthy. These values must be determined by each laboratory according to the respective medical teams, in concordance with their acquired experience. The bibliography presents the approved lists of critical values, that place the CK activity value above 1,000 U/L in this alarm point. Given that the reference values obtained in this paper do not exceed the upper limit of this value, this study group considers that it should not be modified[28-30].

Conclusions

From the analysis carried out on CK serum activity results among male and female elite athletes, specific reference intervals were obtained that differ from those generally used in the laboratory for healthy subjects without the stimulus of physical activity. The limits of the intervals obtained are higher and differ by gender, higher in men than women.

These differences suggest the need to review the reference intervals used for athletes, and each laboratory has its own values, to be able to interpret them correctly. We suggest setting alarm values, to improve assessment of results, from which other diagnostic alternatives should be considered that might influence them. The value of the biochemical expertise, in the knowledge and interpretation of the studies performed on athletes within their training and health control plan is relevant when organising the distribution of workloads, preventing lesions and ensuring their healthcare.

Conflicts of interest

The authors declare that there were no conflicts of interest.

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