



Changes of Cardiac Biomarkers in Ultramarathon Runners

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Abstract

Introduction: The aim of this study is to examine and evaluate the effects of long-term stress on muscle and heart biomarkers after completing a 100 km ultramarathon. **Material and Methods:** Venous blood samples of nine runners (average age 38.8±10.2 years), who successfully finished a 100 km ultramarathon at an elevation of 3130 m, were examined before the start, at the finish line immediately after the run, one day after the run and then after 5 and 10 days. Clinical, laboratory, and somatometric data were obtained from all measurements, and biomarkers: aspartate aminotransferase (AST), cortisol (COR), troponin T (cTnT), creatine kinase (CK) and C-reactive protein (CRP). Also, their training experience and ultramarathon experience was monitored. Discovered values were further analyzed with the use of t-test a ω^2 ($\omega^2 \geq 0.1$), and Spearman's rank correlation coefficient (r) at the significance level of $p < 0.05$. **Results:** The average finish time of the runners was 13:55:40 (min: 12:12:35, max: 16:52:02). After finishing the ultramarathon, runners showed an average weight loss of 2.4 kg ($p < 0.05$). The results show that hematological changes were caused by physiological stress and long-term physical load. The values of all monitored biomarkers showed a significant exceeding of the normal values immediately after the race in 8 competitors out of 9. The values of cTnT showed an increase of more than 50 % (pre-race: 8.2±2.3, post-race: 34.22±25.9 ng/l, max=98 ng/l). After 24 hours, however, this condition had returned to the normal values for all participants. The results show that the AST hepatic enzymes significantly correlated with the training experience ($r = -0.41$, $p = 0.043$), the total number of kilometers run per year ($r = -0.45$, $p = 0.04$) and the achieved finish time ($r = 0.67$, $p = 0.001$). At the same time, athletes who had the best finish time achieved lower CRP values ($r = 0.74$, $p = 0.023$) and cTnT values ($r = 0.49$, $p = 0.040$). The study found that the competitors who had the longest experience with ultramarathon had the lowest cTnT ($r = 0.44$, $p = 0.050$). **Conclusion:** Long-term physical stress is associated with metabolic and cardiovascular changes. Blood abnormalities found in our study suggest that due to long-lasting extreme stress, heart exhaustion may occur. However, these changes did not last long and after a few days they returned to the normal values for all runners.

Keywords: extreme terrain running, heart, physical load, troponin

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INTRODUCTION

More and more scientific observations show that running can also contribute to prolonging “youth” into the late age and therefore can prolong life [1]. Over the last few years, in many countries around the world, running has grown in all its forms on a mass-scale. Ultramarathon or similar forms like skyrunning are developing very dynamically. More than 800 official ultramarathons are currently held around the world, with almost 700 in Europe and North America [2]. According to the Ultrarunning association in 2014 almost 100,000 runners competed in ultramarathon races in the USA [3].

The question is whether such a long-term extreme physical load still has a positive effect on the health of runners. It has been proven that despite the fact that physical activity can positively influence the function of the cardiovascular system, running is a physical stress for the organism. According to some studies [4–5], an increase in cardiac biomarkers, which are indicators of cardiac myocardial damage, already occurs during short-term intensive exercise. Increase in levels of cardiac biomarkers occurred in up to 75 % of the runners studied. Further studies focused on the half marathon or marathon even account for up to 100 % occurrence of elevated troponin levels [6–7]. This is also confirmed other researcher [8,9], who say that up to 94 % of athletes who have finished a marathon-run have shown an increase in highly sensitive troponins after a run.

The question remains: what is the situation in regard to ultramarathon races? Ultramarathon running is in 98 % of the cases carried out within the aerobic mode and a runner goes into a so-called “persistent state,” in which it is possible to run for a relatively long time [10–11]. The speed which the ultramarathon runner is able to develop during this period depends on the overall body fitness. At the same time, it is necessary to maintain a regular supply of energy sources and minerals in order to avoid the disruption of the internal environment of the organism.

In the last 10 years, the issue of the influence of the ultramarathon on cardiac biomarkers has been studied several times. The first studies were used to evaluate Creatine kinase or C-reactive protein. The vast majority of studies used troponin T for their purposes. Recent studies evaluated their analyses using troponin I. In total, 189 runners (184 men and 5 women) were examined. The results of these studies indicate that the increase in cardiac biomarkers above the normal ranges from 10–100 % and elevated cardiac biomarkers appeared in almost all studies. Only one study [12] showed no increase in troponin levels. Overall, the blood troponin levels increased in over 50 % of runners competing in the ultramarathon [13–18]. In all cases, these elevated values returned to normal within 48 hours of recovery.

Based on the above research, it appears that in the ultramarathon the risk of heart damage is much lower than in the marathon due to a much lower involvement of anaerobic modes during performance. However, these findings need to be further refined and therefore the aim of this study is to help clarify this issue. At the same time, the aforementioned studies did not address the possible influence of other factors affecting the runner’s performance. Therefore, in our research we will also focus on the correlation of cardiac biomarkers with performance factors such as age, ultramarathon experience, the training activity of runners, etc. The aim of this study is to examine and evaluate the effects of long-term stress on muscle and heart biomarkers after completing a 100 km ultramarathon.

MATERIAL AND METHODS

Participants

We invited nonprofessional male runners participating in the ultramarathon at 100 km with an elevation of 3130 m to participate in our study. From 21 male athletes recruited, 9 runners agreed to take part in our study. They habitually trained about 240 km a month on average. Before the start of the run, all the runners were assessed for body composition using a model Bodystat 1500 MDD (Bodystat Ltd., British Isles), completed a survey on their medical and training history, and signed an informed consent to participate in the study after being informed about the experimental procedures and risks involved. The study protocol was designed in accordance with the Declaration of Helsinki and was approved by the Research Ethics Committee of the University – No. 1/2017/07.

Study protocol

The contestants participated in a 100 km off-road event, which was located near Ústí nad Labem. Weather conditions at the start of the race (5:00 AM) were good (15 °C and 53 % relative humidity) and did not significantly deteriorate during the race (18 °C and 54 % relative humidity). Participants were allowed free (unrecognized) food intake (sandwiches, biscuits, pasta, bananas, and energy bars of carbohydrates) and liquids (water, sports drinks, soft drinks, and broth). Samples of venous capillary blood were taken 3 hours before the start of the competition, immediately after the run-in to the target. Within 1 hour of taking the blood, the samples were carried to a nearby clinical laboratory for processing and analysis. Additionally, the samples were analyzed in the clinical laboratory 24 hours after the run, and 5 and 10 days after the end of the run. Two weeks before the run, the participants were subjected to physiological testing to assess their maximum oxygen uptake (VO_2max) and their lactate threshold (LT). As ultraendurance runners mostly rely on high volume training at low to moderate intensities, with no speed training targeted at the development of motor coordination required for fast running, the test consisted of a combination of varying treadmill speeds and grades. The test was performed on a model HP Cosmos 3005 treadmill (HP Cosmos, Germany), beginning at 10 km/h and at a 5° inclination. The treadmill speed was increased by 1 km/h every 1 min until volitional fatigue was reached. During the test, heart rate, minute ventilation (VE), oxygen uptake (VO_2), and expired carbon dioxide (CO_2) were continuously measured using a model MetaLyzer 3B-2R stationary spirometer (Cortex, Leipzig, Germany) in the breath-by-breath mode. Fingertip capillary blood samples for the assessment of lactate (LA) concentration were drawn at the end of the run. Using these data, an individual LT was estimated by the D-max method [19].

Blood sampling

In order to establish changes in the blood composition of the subjects before the ultramarathon and after the race's finish, blood was collected from the antecubital vein in accordance with the criteria of the CLSI Guidelines (Clinical and Laboratory Standards Institute). As for creatine kinase (CK), CK-MB, cardiac troponin T (cTnT), and high sensitive C-reactive protein (CRP), blood was collected using vacutainer SST tubes (BD Vacutainer Serum Separator Tube, USA) including gel and agent for promoting blood coagulation, centrifuged at 3,400 rpm for 10 min, separated from serum, and frozen at -70 °C to be measured later.

Statistical analysis

Results are expressed as mean, median, minimum, and maximum values and show standard deviations. To assess the impact of the race on the data obtained in the two predetermined moments (pre- and post-race), the Student's t-test was used for paired samples. The variables' normality was analyzed by the Shapiro-Wilks test. The effect size was evaluated using omega ω , which deals with the problem of η^2 distortion [20]. We consider differences with a value of $\omega^2 \geq 0.1$ to be significant [21]. Spearman's rank correlation coefficient (R) was calculated to assess associations between variables; differences between R values were tested by a single-sided test. We also used a multiple correlation coefficient for individual biomarkers. Values over $r \geq 0.4$ were considered significant [21]. In all cases, a $p < 0.05$ was considered to be significant. All statistical analyses were run using the Statistica 7.1 software package (StatSoft, Inc.).

RESULTS

All volunteers included in the study were healthy with an absence of high BP, diabetes, cardiovascular diseases, and liver disorders. Anthropometric and physiological characteristics of the participants are presented in Table 1.

Upon completion of the ultramarathon, runners showed a weight loss of 2.4 kg on average ($p < 0.05$). The average time measured at the finish line was 13:55:40 (min: 12:12:35, max: 16:52:02). When evaluating the observed biomarkers, we found that a significant exceeding of the normal values of all monitored biomarkers immediately after the race occurred in 8 out of 9 competitors (Table 2). When comparing pre-race and post-race values, we found that for all monitored biomarkers there was

a statistically significant difference and a significant effect size in pre-race and post-ultramarathon values ($\omega^2 \geq 0.1$) (Table 3).

In Table 4 we focused on an evaluation of the correlation between the studied biomarkers and performance factors such as age, experience with the ultramarathon, the training activity of runners, etc. From the results it is evident that liver enzymes AST correlated significantly with the training experience ($r = -0.41$, $p = 0.043$), The overall amount of kilometers run over a year ($r = -0.45$, $p = 0.040$) and the finish time achieved ($r = 0.67$, $p = 0.001$). At the same time, runners who had the best finish times exhibited lower values of CRP ($r = 0.74$, $p = 0.023$) and cTnT ($r = 0.49$, $p = 0.040$).

Table 1. Anthropometric and performance characteristic of participants

Variable	Mean \pm SD	Min-Max
Age [years]	38.3 \pm 10.0	26-53
Weight [kg]	70.9 \pm 6.3 (68.5 ^w \pm 5.1)*	62-84
Height [cm]	178.4 \pm 6.2	168-189
Body mass index [kg/m ²]	22.2 \pm 1.1	20.5-23.5
Body fat [%]	18 \pm 6.7	11.1-24.3
Peak oxygen uptake [ml/kg/min]	65.2 \pm 4.6	58.4-72.1
Weekly covered distance [km]	239.7 \pm 75.4	150-400
Training history [years]	9.7 \pm 4.3	5-16
Yearly covered distance [km]	3022.2 \pm 677.8	2300-4500
Race time [hours]	13:55:40 \pm 3:00:06	12:12:35-16:52:02
High BP [%]	0	-
Diabetes [%]	0	-
Cardiovascular diseases [%]	0	-
Liver disorders [%]	0	-

SD - standard deviation, ^w - body weight after the ultramarathon; * - $p < 0.05$

Table 2. Biomarker values during the ultramarathon run

Variable	URL	Pre-race	Post-race	24 hours post-race	5 days post-race	10 days post-race
AST [μ kat/l]	0.75	0.59 \pm 0.2	2.90 \pm 3.24	2.68 \pm 2.24	1.42 \pm 1.39	0.66 \pm 0.17
CK [μ kat/l]	2.9	5.06 \pm 5.2	115.4 \pm 173.5	53.5 \pm 57	6.93 \pm 6	3.97 \pm 2.1
CRP [mg/l]	5.0	1.49 \pm 0.8	10.37 \pm 8.5	23.18 \pm 17.2	5.21 \pm 1.5	1.71 \pm 1.1
COR [nmol/l]	477	402.0 \pm 172.7	941.1 \pm 309.9	441.8 \pm 162.3	388.2 \pm 168.2	390.2 \pm 202.1
cTnT [ng/l]	14.0	8.2 \pm 2.3	34.2 \pm 25.9	6.6 \pm 3.2	-	-

AST - aspartate aminotransferase; CK - creatine kinase; CRP - C-reactive protein, COR - cortisol; cTnT - troponin T

Table 3. Biomarker changes during the ultramarathon race

Variable	Pre-race vs post-race			Pre-race vs 24 hours post-race			Pre-race vs 5 days post-race			Pre-race vs 10 days post-race		
	t-test	p	ω^2	t-test	p	ω^2	t-test	p	ω^2	t-test	p	ω^2
AST	-2.24	0.056	0.31	-2.86	0.021	0.44	-1.71	0.125	0.18	-0.96	0.364	-0.01
CK	-1.96	0.085	0.24	-2.72	0.026	0.42	-1.59	0.149	0.15	0.98	0.357	0.00
CRP	-2.39	0.009	0.34	-3.39	0.004	0.54	-2.94	0.018	0.46	-0.45	0.659	-0.10
COR	-2.12	0.066	0.28	4.66	0.001	0.70	3.78	0.005	0.60	4.71	0.002	0.70
cTnT	-3.20	0.012	0.51	1.44	0.186	0.11	NM	NM	NM	NM	NM	NM

AST - aspartate aminotransferase; CK - creatine kinase; CRP - C-reactive protein, COR - cortisol; cTnT - troponin T; NM - not monitored

Table 4. Correlation of biomarkers measured after the race with the training load and performance factors during the race

	AST post-race		CK post-race		CRP post-race		COR post-race		cTnT	
	r	p	r	p	r	p	r	p	r	p
Age [years]	0.00	0.998	0.08	0.846	0.49	0.184	-0.23	0.556	-0.42	0.256
BMI	0.36	0.343	0.29	0.445	0.38	0.311	-0.04	0.912	0.43	0.250
VO ₂ max [ml/kg/min]	-0.26	0.143	-0.14	0.080	-0.11	0.290	-0.25	0.092	-0.15	0.560
Weekly covered distance [km]	0.03	0.942	0.08	0.846	-0.27	0.489	-0.33	0.393	-0.12	0.785
Training history [years]	-0.41	0.043	-0.23	0.564	-0.39	0.052	-0.36	0.450	-0.44	0.050
Yearly covered distance [km]	-0.45	0.040	-0.16	0.250	-0.44	0.030	-0.21	0.174	-0.29	0.174
Race time [hours]	0.67	0.001	0.02	0.954	0.74	0.023	0.03	0.935	0.49	0.040

AST – aspartate aminotransferase; CK – creatine kinase; CRP – C-reactive protein, COR – cortisol; cTnT – troponin T

DISCUSSION

It is generally stated that running is one of the simplest forms of human motion and the most natural way to experience psychological and physical fatigue. Over the last few decades, running has generally been recognized as an effective preventive measure against the diseases of civilization. Running is a permanent part of a healthy lifestyle, strengthens the heart and blood circulation, and reduces the resting heartbeat frequency and increases the ability to tolerate more physical stress. On the other hand, there are some critical points which should be observed in relation to health prevention of athletes, when physical activity is too intensive. In our study, we evaluated the effects of long-term stress on muscles and heart biomarkers after completing 100 km ultramarathon. When comparing pre-race and post-race values, we found that for all monitored biomarkers, there was a statistically significant difference and a significant effect size in pre-race and post-ultramarathon values. Our results confirmed conclusion by Kłapcińska et al. [14]. The aspartate aminotransferase was five times higher values measured after the completion of the ultramarathon and pre-race values. This value remained the same even the day after the race and it is evident that in 44 % of cases it was affected by physical stress during the ultramarathon. Creatine kinase showed a significant increase from the pre-race value of $5.06 \pm 5.2 \mu\text{kat/l}$ to $115.4 \pm 173.5 \mu\text{kat/l}$ after the race (Table 3). Similar results [22] (mean creatinine and cortisol increased significantly by 30.5 % and 291.4 %), were found with cortisol where the value was at an increased level even 10 days after the race. In all of the measurements we found a statistically significant difference and a significant effect size between the pre-race value and the post-race value. On the other hand, our results depart from the ones presented by Da Ponte et al. [22] with the C-reactive protein biomarker, which examined the runners during an uphill run with the load time of only around 4 hours. C-reactive protein value in our research was significantly increased 5 days after the race (Table 3).

Also, when analyzing the dynamics of troponin T, we found that values before the ultramarathon and after the ultramarathon showed a significant increase due to extreme stress. When comparing absolute values, it changed from the value of $8.2 \pm 2.3 \text{ ng/l}$ before the race to a value of $34.2 \pm 25.9 \text{ ng/l}$ after the race. The significant difference was 51% caused by the extreme load intensity (Table 3). Similar results were also reached by studies such as [15–18, 23]. It is evident and proven by a number of studies that high-intensity or long-term physical load causes damage to skeletal muscles, with damage to the cellular membrane and a release of intracellular substances such as Creatine kinase, C-reactive protein and also troponin T into the blood circulation [12,24–25].

In our study, the level of elevated troponin of runners significantly correlates with age, experience, and the training distance of runners. Similar results were obtained by Li et al. [16]. On the contrary, Da Ponte et al. [22] reached the opposite results, and did not detect a significant dependence between an elevated troponin level and monitored parameters such as age, BMI, VO₂max, training experience, and race time.

We also found that those competitors who had the longest experience with the ultramarathon had the lowest levels of troponin T. This fact has been proven by research [26]. At the same time, we

have proven, in confirmation of research by Eijsvogels et al. [27], that the finish time in the ultramarathon significantly correlated with the level of troponin T in the blood (Table 4.), and we can confirm that there is a correlation between increased cardiac biomarkers and load time. However, some authors have come to other conclusions and say that the effect on the more significant release of cardiac biomarkers is due to the intensity of the load rather than its duration [4–5,28] This fact is confirmed for example by Kim et al. [17], who found that elevated troponin T values for the ultramarathon were lower when compared to shorter loads such as in the marathon. Recently, It has been documented that the dynamic of secretion cardiac biomarkers were differentially regulated during the 24-hour ultra-marathon and both exercise duration and intensity may play a crucial role in cardiovascular adaptive mechanisms and cause a higher risk of cardiac stress in ultra-marathoners [29].

We are aware that our work has certain limitations. The first fact is that we were unable to track the intensity of the load with a recording device during the race. The monitored ultramarathon runners were not used to using a heart-rate band, and therefore we were forced to withdraw our plan to use them. The second limitation is the fact that we did not monitor the amount of fluids consumed during the races and the contribution of the circulatory and thermal components to the physiological strain in runners [30]. The final limitation of our study is the size of the research sample, which may have been reflected in the evaluation of correlations between the monitored variables. Despite these limitations, we believe that if the physical load is long-lasting and of medium intensity, in runs like the ultramarathon, there is a significant increase in biomarkers that indicates the presence of skeletal and cardiac muscle damage regardless of age, fitness status, and performance.

CONCLUSION

Long-term physical load is associated with metabolic and cardiovascular changes. Blood abnormalities found in our study suggest that heart fatigue due to long-lasting extreme physical loads may occur. However, these changes did not last long and after a few days these indicators returned to normal values for all runners. The long-term effect of these changes is still unknown. Increased levels of cardiac biomarkers after exercise are affected by training experience and performance level.

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