Effects of Sixty-Minute Race-Pace Running on Cardiac Stress Biomarkers in Recreational Distance Runners

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ABSTRACT

Sudden cardiac death (SCD) in athletes is generally rare, but a serious complication of cardiovascular events during exercise. Although regular intensive physical exercise is thought to be a key to a healthy life, unsuspected pathologies might lead to SCD during or after physical activity. Cardiac dysfunction and elevated cardiac markers have been reported after prolonged exercise. We sought to clarify the cardiac marker levels and hydration status in healthy, middle-aged male subjects for 24 hours after running sixty-minute at race-pace. The participants were 47.4±1.7 years old, had peak oxygen consumption of 47.1±1.2ml/kg/min, and regularly running 70.5±6.4km/week. Blood biomarkers were performed before, immediately after, at the fourth and twenty-fourth hours after running. Compared to initial values, creatine kinase (before:161.2±22.5U/L, 24 hours after:411.9±139.7U/L, $p<0.001$) and CK-MB (before:4.3±0.7ng/ml, 24 hours after:10.1±3.0ng/ml, $p<0.001$) were significantly elevated immediately after running and remained significantly high for 24 hours. In addition, Troponin-I (before:5.0±1.1ng/l, 4 hours after:81.5±29.9ng/l, $p<0.001$) and NT-proBNP (before: 31.2±5.3pg/ml, immediately after: 64.4±8.5pg/ml, $p<0.01$) were significantly elevated immediately after running and returned to baseline levels in 24 hours. The sixty-minute running caused significant dehydration, but athletes were rehydrated at the 4th hour in their voluntary hydration behavior. As the individual data were analyzed, it was interesting to see that some of the athletes had critical biomarker levels without any cardiac symptom. Our findings indicate that race-pace sixty-minute running may induce a possible transient silent myocardial injury in apparently healthy master runners. Detailed pre-participation screening of these athletes may be necessary to reduce the risk of SCD.

KEYWORDS: CK-MB, Creatine kinase, Marathon run, Sudden cardiac death, Troponin-I.

INTRODUCTION

In recent years, it has been observed that there has been an increase in participation in sports. However, it is also stated that some cardiac complications and even sudden cardiac death (SCD) may occur during or after physical activities such as marathons and ultramarathon running. The actual frequency of SCD during exercise is unknown, but some studies reported that it has increased from around 1:300.000 in the 1990s to 1:50.000 in 2021[1, 2]. Three-fourths of the SCDs occur during or just after physical activity, and most of these events develop in adults over 35 years old[3].

Exercise is generally accepted as cardioprotective, and most long-distance runners do not complain about cardiac symptoms either during or after training. However, endurance activities may be associated with severe complications such as atrial fibrillation, arrhythmogenic right ventricular cardiomyopathy, and hypertrophic cardiomyopathy. Even the life-threatening events are rare, long-distance running may lead to right ventricular dysfunction, inflammation, and the release of cardiac damage markers in endurance athletes[4]. Monitoring the stress experienced by recreational distance runners due to training may be important to interpret cardiac complications that may occur in the chronic period. With this in mind, the evaluation of the stress that runners encounter during training may have particular importance.

Generally, recreational distance runners train for 50 to 60 km in their weekly routine and run approximately for an hour daily [5]. In addition to the intensity of the exercise during the training, changes in the body fluid balance may affect the cardiac strain, especially in a hot environment [6]. Nevertheless, hourly fluid loss can exceed 1 liter in physical activities performed in relatively cooler environments[6]. Training intensity, together with dehydration, may cause serious cardiac complications in athletes, especially with the presence of an underlying cardiac disease. Thus, evaluating the changes in cardiac markers and dehydration to understand the physiological stress level during training for these runners, and this issue needs clarification to guide long-distance runners.

Elevation of cardiac markers may indicate asymptomatic ischemia, even without any symptom and ECG abnormalities[7]. The cardiac strain is frequently determined by measuring plasma total creatine kinase (CK), creatine kinase muscle-brain fraction (CK-MB), and the ratio of CK-MB to total CK (the CK index) with Troponin-I concentrations[8]. Besides that, N-terminal prohormone of brain natriuretic peptide (NT-proBNP) is released from myocytes as a result of increased ventricular wall stress, hypoxia, and myocardial infarction[9-11], which is used as a cardiac marker to show structural abnormalities such as filling dysfunction or right ventricle overload during prolonged exercises [12]. After a training period, following the plasma level of these cardiac markers for 24 hours may be

valuable to detect possible non-symptomatic cardiac complications in recreational distance runners, which was not studied previously.

With this in mind, in this study, we aimed to study the effect of a 60-minute running session on the cardiac biomarkers, dehydration, and ECG that may pose a risk for cardiac complications within the following 24 hours.

METHODS

Participants

Twenty-one male runners (age: 47.4 ± 1.7 years, height: 171.0 ± 1.3 cm, weight: 73.3 ± 1.7 kg) were enrolled in this study. All tests were carried out in the morning hours with a constant temperature of 23 ± 1°C in Cukurova University Wellness and Sports Sciences Research Centre. The study was in accordance with the Declaration of Helsinki, with the approval of the Cukurova University Faculty of Medicine Clinical Researches Ethics Committee no. 2018/83. The procedures and purposes of the study were explained to all participants in detail, and informed consent forms were obtained.

Preliminary Measurements

A detailed medical examination of the runners was performed by the cardiologists, and runners with any suspicious situations were excluded from the study. The runners included in the study were all healthy up to cardiologist consultations. The runners remained in the supine position for approximately 10 min for resting HR recording before the medical examination. The blood pressure measurements, pulmonary function tests (slow vital capacity (SVC), forced vital capacity (FVC), maximal voluntary ventilation (MVV); Quark b^2, Cosmed, Italy), and ECG recordings (Quark T12x, COSMED, Italy) were performed on the first visit day of the laboratory. Any disease and drug usage were accepted as exclusion criteria. Subjects were instructed to refrain from exercise for all laboratory visit days in the preceding 24 hours. Anthropometric measurements were performed to determine the body composition. Body weights (BW) were measured on a scale with a sensitivity of 0.02 kg (Kurdaklar Baskül, Turkey), and height was measured by a stadiometer (Sport Expert, Turkey) with a sensitivity of 0.01 cm while standing in an upright position. Upper thigh, calf, and forearm circumference measurements were made with non-elastic tape. Subscapular, triceps, biceps, forearm, abdominal, pectoral, suprailiac, thigh, and calf skinfold thicknesses were determined by using a skinfold caliper (Holtain, England). Siri formula was used to calculate body fat percent values and Martin formula for muscle percent values [13, 14].

Cardiopulmonary exercise test (CPET)

The maximal cardiopulmonary exercise test was applied by indirect calorimetry (Omnia, Cosmed, Italy) to determine performance status on the first visit day. The treadmill (HP Cosmos, Nussdorf - Traunstein, Germany) started with a 1% incline and 6 km/h speed and was adjusted to increase the speed automatically by 1 km/h every minute. Heart rate was recorded simultaneously (Cosmed, Italy). The test completion criteria were accepted as reaching to 90% of the maximal theoretical heart rate, a plateau in oxygen uptake, a non-protein respiratory quotient (RQ) value above 1.15, or the participant's verbal declaration that could no longer continue[15]. The anaerobic thresholds (AT) were calculated by the V-slope method [16]. Athletes run at the running speed corresponding to the oxygen uptake value at the midpoint of maximal VO_2 and the VO_2 at the anaerobic threshold. Athletes' running paces in the 60-minute running test were individually derived from the data in the CPET. Oxygen uptake values rather than heart rate was used to calculate the individual running speed.

Sixty-minute running test

On the second visit day, the participants performed a 60-minute running test on a treadmill (LifeFitness CLST, USA) at a determined running pace. Before, immediately after, at the 4^{th} and 24^{th} hours following the running trial, venous blood samples were taken from the antecubital vein to analyze cardiac markers (total CK, CK-MB, NT-proBNP, Troponin-I) and hemogram. Capillary blood samples from the fingertip were taken for blood glucose and lactic acid level measurements (Biosen Lactate and Glucose Analyzer, EKF Diagnostics GmbH, Germany) before and immediately after sixty-minute running test. Body weights were measured with only underwear. The runners were free to consume only pure unsweetened water, but the amount of water consumption was weighed and recorded. A measuring cup was given to the participants who needed to urinate, and the urine volume was determined. The warm-up session started with walking at 6km/h and 1% incline on the treadmill, and the participants voluntarily increased the speed up to their personal running speed. At the 6^{th} minute, the treadmill had stopped, and all runners stretched for two minutes. During the sixty-minute running test, participants were not allowed to exceed the specified running pace. The individuals who had trouble keeping running pace were allowed to reduce running speed gradually to the AT pace by the supervision of the test administrators. The running speed changes and duration were recorded to calculate the average running speed and distance. The test termination before sixty minutes was accepted as an exclusion criterion for this study. The heart rates were recorded continuously (Garmin Forerunner 305) during the test, and the fatigue levels were questioned by the Borg Scale (6-20 scale) every 10 minutes. Immediately after the sixty-minute running test, ECGs were recorded, and blood samples were taken to evaluate hematological parameters. Runners were reweighed to determine post-running body mass by wearing only their underwear. Changes in body weight and the amount of fluid consumed and urinated were calculated.

Blood sampling

A professional nurse performed all the blood sampling. The samples were withdrawn from the antecubital vein (10 mL) directly to anticoagulant-containing (EDTA) vacutainer tubes for hemogram and non-anticoagulant containing vacutainer tubes (BD, US) for blood biochemistry analysis. The EDTA tubes were inverted gently to mix the blood with the anticoagulant. The samples were transported to the Cukurova University Faculty of Medicine Balcalı Hospital, Central Laboratory (certified by the Joint Commission International) in an insulated cold box. Hemogram, total CK, CK-MB, and Troponin-I kits were provided by Beckmann Coulter (Beckmann Coulter, US), and the NT-proBNP kit by Roche (Rotkreuz, Switzerland). The kinetic enzymatic method (Beckmann Coulter AU5800, US) was used for total CK, and the chemiluminescence method was used for CK-MB and Troponin-I (Beckmann Coulter DXI600, US, and Beckmann Coulter DXI600, US, respectively) measurements. NT-proBNP was measured by the electrochemiluminescence method (Maglumi2000, Shenzen, China). White blood cells, red blood cells, hemoglobin, and hematocrit were measured photometrically by an automated analyzer (Beckmann Coulter DXH800, US).

Capillary blood samples were withdrawn from the fingertip. Twenty microliters of blood samples were used to analyze immediately with an automatic analyzer (Biosen Lactate and Glucose Analyzer, EKF Diagnostics GmbH, Germany).

Statistical analysis

SPSS version 21 for Windows was used for statistical evaluations. The distribution of the data was evaluated with the Shapiro-Wilk test. Paired t-test or repeated measures of ANOVA followed by Bonferroni *post hoc* were used for normally distributed data. Friedman and Wilcoxon tests were used for data that did not normally distribute. The confidence interval was set as 95%. Spearman's correlation coefficients were used to examine the relations between variables. Values were presented as mean ± standard error. Statistical significance was set at $p<0.05$.

RESULTS

Twenty-one male runners were enrolled in the study. The participants were training regularly for 11.8±2.0 years and running 70.5±6.4 km per week. Two of the 21 runners could not finish the 60-min running test due to fatigue and excluded from the study. The individual running pace was 86.44±0.91 % of the maximal oxygen uptake value, and this pace corresponds to 90.3±2.6% of the heart rate reserve. The physical characteristics, anaerobic threshold, and maximal running values during CPET of the runners were presented in Table-1 and Table-2, respectively.

188 Insert Table-1 here.

189 Insert Table-2 here.

190

191 The average running speed was determined as 12.4±0.3 km/hour and found to be strongly correlated
192 with the last half marathon average running speed (12.7±1.7 km/hour) declared by the runners
193 ($r=0.846$, $p<0.01$). During the sixty-minute running test, participants reduced their pre-determined
194 running pace, and the average running speed was calculated as 11.5±0.4 km/h. All of the individuals
195 began to run above AT, and none of them finished below the AT level. The maximal and mean HR
196 values were 172.8±2.9 beats/min and 162.0±12.6 beats/min, respectively. The average HR values in
197 each 10 minutes during 60-min running were evaluated. The average HR for the first 10-min was
198 157.4±3.8 bpm, which was estimated as 155.8±2.8 bpm from CPET data. The average HR values
199 were increased during running, and the last 10-min average value was 168.4±3.4 bpm (significantly
200 higher than the first 10-min average value, $p<0.05$, Figure-1). The runners lost 1.6±0.1 kg,
201 corresponding to 2.2±0.2% of their body weight, and consumed 231.6±40.6 ml of water during
202 running. The volume of urine collected from the participants between the body weight measurements
203 were 57.9±22.2 ml. Following the sixty-minute running test, runners' body weight reduced
204 significantly from 73.2±1.7 kg to 71.5±1.7 kg ($p<0.05$). Pre-test lactate values (2.3±0.2 mmol/L)
205 increased significantly ($p=0.00$) following the sixty-minute running test (5.4±0.6 mmol/L). Runners'
206 perceived exertion for the first ten minutes was 9.2±0.5 and increased to 11.2±0.6 during the test, and
207 this difference was found significant ($p<0.001$). Also, post-test glucose values were increased
208 significantly compared to pre-test values from 5.0±0.2 mmol/L to 6.6 ± 0.6 mmol/L ($p=0.00$). In
209 addition to that, there had been no pathological ECG recordings before, after, and at the 4^{th} and 24^{th}
210 hours following the sixty-minute running test.

211 Insert Figure-1 here.

212 Cardiac marker levels before, after, and at the 4^{th} and 24^{th} hours following the sixty-minute running
213 test are shown in Figure-2 and Figure-3. Before the sixty-min running test, total CK levels were below
214 the cut-off value (170 U/L) except for four runners. The pre-test average total CK value increased
215 significantly ($p<0.001$) following the sixty-minute running test and remained high for 24 hours
216 ($p<0.001$). Moreover, 24^{th} hour total CK values were significantly higher than the post- and 4th-hour
217 values ($p<0.05$). The number of runners with total CK values above the cut-off level immediately
218 after running test and at the 4^{th} hour was eleven and increased to fourteen at the 24^{th} hour. Pre-, post-
219 , 4^{th} and 24^{th} hour average total CK values were measured as 161.2±22.5 U/L, 222.1±31.9 U/L,
220 274.1±57.8 U/L and 411.9±139.7 U/L, respectively (Figure-2A).

Plasma CK-MB values had a similar pattern as total CK. Before running, CK-MB average value was between the normal range (0.97-4.94 ng/ml). The individual data showed that six runners' CK-MB values were above the reference value before the test, and after 24 hours, 13 runners' CK-MB values were higher than the cut-off value (highest value: 61.1 ng/mL). The average CK-MB value increased significantly ($p<0.001$) immediately after the sixty-minute running test and stayed high for the following 24 hours. Besides that, the 4th and 24th hour average CK-MB values were significantly higher than immediately after exercise ($p<0.05$ and $p<0.05$, respectively). Pre-, post-, 4th, and 24th hour average CK-MB values were measured as 4.3±0.7 ng/ml, 5.8±0.9 ng/ml, 7.7±1.6 ng/ml, and 10.1±3.0 ng/ml, respectively (Figure-2B).

Eight runners' CK index value was higher than the cut-off limit (2.5%) before the running test. Immediately after running CK index did not show any significant difference; however, the 4th hour CK index value was significantly higher than pre- and post-exercise ($p<0.05$). At the 24th hour, eleven runners' CK indexes were above the cut-off value. Pre-, post-, 4th and 24th hour average CK index values were calculated as 2.7±0.3%, 2.7±0.3%, 2.9±0.3% and 2.8±0.3%, respectively (Figure-2C).

Insert Figure-2 here.

Before the running test, the Troponin-I average value was between the normal range (10-40 ng/l). Individual data had shown that none of the runner's initial Troponin-I was above the reference value. Following the running test, plasma Troponin-I values increased significantly for 24 hours. However, the most significant changes were detected at the 4th hour ($p<0.001$), and seven runners' Troponin-I levels increased above the cut-off value (highest value: 539ng/L). Even though the difference between the initial and 24th hour average Troponin-I value was significant, the 24th hour average value was below the cut-off level. Pre-, post-, 4th and 24th hour average Troponin-I were measured as 5.0±1.1 ng/l, 11.5±2.4 ng/l, 81.5±29.9 ng/l and 15.1±3.2 ng/l, respectively (Figure-3A).

Before running test, the NT-proBNP average value was lower than the reference value (125 pg/ml). Analyzing the individual data, one runner's NT-proBNP was elevated immediately after the sixty-minute running test. During 24 hours, the average NT-proBNP did not exceed the reference value. Pre-, post-, 4th and 24th hour average NT-proBNP values were measured as 31.2±5.3 pg/ml, 64.4±8.5 pg/ml, 50.6±6.5 pg/ml and 49.6±7.1 pg/ml, respectively (Figure-3B).

253 Insert Figure-3 here.

254

255 On the other hand, the correlation between the age of the runners and the pre-exercise NT-proBNP
256 values were significant ($p<0.05$, $r=0.510$). In addition to that, the correlation between the initial and
257 post-race NT-proBNP values were significant ($p<0.001$, $r=0.936$) (Figure-3C and Figure-3D,
258 respectively).

259 Hemogram and blood plasma volume changes are presented in Table-3. Post-exercise hemoglobin,
260 hematocrit, and red blood cell values were significantly higher than pre-, fourth, and twenty-fourth
261 hour measurements. On the other hand, post-exercise and 4^{th} hour white blood cell numbers were
262 significantly higher than pre- and 24^{th} hour measurements. The sixty-minute running test significantly
263 reduced the plasma and total blood volumes immediately after exercise by 6.2±1.2 % and 3.6±0.7 %
264 compared to pre-exercise, respectively ($p<0.001$). The plasma and total blood volume loss recovered
265 at the fourth hour after the sixty-minute running test.

266

267 Insert Table-3 here.

268

269 **DISCUSSION**

270 In our study, the athletes reached the maximum HR values at the last minutes of 60-minute running.
271 It was determined that the cardiac damage markers were increased following this running trial, and
272 CK and CK-MB concentrations did not return to normal levels in the following 24 hours. It was also
273 important to see that sixty minutes running at room temperature caused significant dehydration; even
274 the participants were allowed free water consumption.

275 The elevation of HR values during prolonged running may be explained by cardiac drift [17]. Athletes
276 may ignore the changes in heart rate during their daily training or competition to keep the pace
277 constant. Since the primary goal of our study was to evaluate the effects of physiological stress that
278 runners had to cope with during their daily training, we did not adjust running speed up to heart rate
279 changes. The athletes should be aware of the risks on the field, such as different environmental
280 conditions, such as high altitude, hypoxia, and cold/hot weather. In the light of these findings, the
281 stress encountered in daily training can create a substantial burden for recreational marathon runners.

282 ***Pre-exercise***

The 24-hour period between the last exercise session and the 60-min exercise test is important to simulate runners' consecutive training, and pre-exercise cardiac marker evaluation is valuable to reflect their daily routine. These runners' ECG data showed no abnormalities, and this finding claims that our participants had no cardiac damage before the study. Even the mean values of all cardiac markers were within the normal range before the 60-min running test; four runners' total CK, six runners' CK-MB, and ten runners' CK index values were higher than reference values. It has been shown previously that, following consecutive training sessions, reduction of CK and CK-MB may take 28 hours to several days to return to the normal range [18, 19]. Moreover, the recovery period after running may take a longer time in masters compared with younger athletes[20]. Most of the cardiac marker data given in the literature express the values following a marathon or ultramarathon events. However, the high marker levels in some runners that we evaluated following the 60-minute running trial, which may simulate runners' daily training activity, indicate that 24 hours of recovery time may be insufficient for these markers to return to reference values. Although within physiological limits, the positive correlation between age and pre-test NT-proBNP values suggests that elderly master runners recover after ventricular loading later than younger ones. Long-term consecutive training sessions may trigger cardiac remodeling, which may induce ventricular fibrotic lesions and ventricular arrhythmias, especially in elderly runners who have trained for a long time[19]. This finding indicates that elderly runners require to be conscious of cardiac strain during their daily routine.

Post-exercise

Exercise-induced elevation of total CK, CK-MB, and CK index values has been shown previously, and our findings are in agreement with the literature [21]. Kobayashi et al. published that total CK peaked at the 24th hour following a marathon run and remained significantly elevated for three days [22]; our findings are important to underline that a single 60-min exercise training session may cause cardiac strain. Following the sixty-minute running test, significant elevation of total CK and CK-MB indicates that consecutive training sessions may accumulate cardiac strain, especially for the runners who initially had higher marker levels. Since following a cardiac strain, the CK and CK-MB values peak in the following hours, post-exercise blood sampling may not be enough to detect cardiac abnormalities, and follow-up sampling may be more valuable. On the other hand, it has been shown recently that heart rate variability measurements may be more practical to detect cardiac strain both in athlete and recreational distance runners [23].

The data published by Kosowski et al. indicated that both NT-proBNP and Troponin-I increase due to exercise, but values below the cut-off limits might not indicate cardiac damage[8]. In our study, even the mean values of Troponin-I and NT-proBNP were within the normal range, they increased

significantly after a 60-min running trial, and one runner's Troponin-I and another runner's NT-proBNP exceeded the reference values. In the literature, it was stated that the increase was limited in half-marathons or marathons [24], the NT-proBNP value could increase to critical levels in longer races such as ultramarathons[18]. In addition, the positive correlation between the initial and post-running NT-proBNP values indicates that older individuals, who had high NT-proBNP values initially, may be more prone to cardiac complications related to ventricular overloading. Even though we did not detect any ECG abnormalities after sixty-minute running exercise, normal ECG data may not be enough to exclude the possible cardiac risks at this time period due to cardiac marker abnormalities [25, 26].

Post-exercise 4th hour

Troponin-I concentration, which was not elevated immediately after exercise, increased significantly at the 4th-hour post-exercise, and 7 of 19 runners' values exceeded the upper reference limit. Although the elevation of Troponin-I concentration after prolonged exercise in elder individuals had been shown previously[8, 27], in our study, no correlation was found between age and Troponin-I concentration. Consistent with Troponin-I concentration changes, the elevation of CK-MB and CK index values strengthens the possibility of cardiac strain. Morville et al. indicated that CK and CK-MB could be elevated after prolonged exercises and maintain high levels for several hours [18], which is in agreement with our findings. Although there were no abnormal ECG signs, the evident elevation of cardiac enzymes in plasma may indicate silent microinfarction, endocardial lesion, or transient cell membrane permeability changes secondary to reversible ischemia [8]. Especially acutely raised Troponin-I, rather than total CK or CK-MB, may represent myocardial necrosis, and episodes of Troponin-I elevation may culminate in abnormal cardiac remodeling and SCD [28].

Post-exercise 24th hour

The highest total CK and CK-MB levels were measured at the 24th hour post-exercise sampling, and most of the runners' values were higher than reference levels. Our findings are in agreement with the previously published data, in which authors showed that total CK and CK-MB might remain elevated significantly for 24 hours or more after endurance exercise [18, 19, 26]. Troponin-I returned to normal range at the 24th hour except for one runner, with normal ECG. Our findings indicate that 60-min training pace running was enough to elevate cardiac markers for 24 hours. Prolonged elevation of these cardiac marker levels may be an indicator of exercise-induced reversible or sub-clinical cardiac damage[18, 20].

Hydration status

It has been shown previously that dehydration may reduce athletic performance and trigger serious cardiac complications after prolonged events, especially in individuals with lower aerobic capacity and in hot environmental conditions [29-31]. Our findings indicate that the sixty-minute running test at room temperature decreased the body weight (2.3±0.2 %), blood volume (3.6±0.7 %), and plasma volume (6.2±1.2 %) and induced significant dehydration. The amount of water loss in a marathon run is approximately 2.5 liters in a cool environment[6]; our 60-min running data is in agreement with the literature. It is also important to underline that; our data showed that ad-libitum water consumption for four hours after running was enough to recover hydration status. Dehydration may reduce ventricular filling and cardiac output during exercise, which restricts tissue blood flow, and thus impacts the whole body's physiological function [32]. Intra- and extracellular fluid content may vary due to dehydration, especially with changes in electrolyte balance. The evaluation of dehydration for these compartments may be more valuable to evaluate the cardiac strain during exercise. Recreational runners generally exercise outdoors, and dehydration, with electrolyte imbalance, may become evident as the ambient temperature increases. Although, in this study, the runners ran indoors and consumed ad libitum water, the 60-min running test was stressful enough to induce significant dehydration. On the other hand, it has been shown that aging may affect sweating responses during exercise together with hydration status and body thermoregulatory function by reducing muscle mass and increasing fat percentage [33]. These changes adversely affect thermoregulatory responses during exercise, and the tolerance to dehydration may decrease due to aging. Exercise-induced dehydration may cause severe complications for aged, untrained individuals requiring special attention [34-36]. We assume that regular training may prevent age-induced changes in their body composition [37], and exercise-induced adaptive changes may enhance thermoregulatory responses in elderly athletes [33]. With this in mind, evaluating hydration levels and taking measures against dehydration may be important to prevent cardiac complications and SCD for recreational distance runners who are training or running long distances.

CONCLUSION

The results of our study indicate that a single bout of running exercise for 60-min may increase HR values and cardiac biomarker levels in regularly training runners. Evaluation of the individual data showed that some of the runners' cardiac markers might elevate dramatically for up to 24 hours, which may be a sign of myocardial ischemia and cause SCD. Even the exercise is generally accepted as cardioprotective; recreational distance runners should be aware of that training may be a double-edged sword. Improving and expanding the cardiovascular screening of athletes, and their continuous education about the possible risks and precautions can prevent serious complications.

The limitations of this study were small sample size, no female runners, and no echocardiography imaging after running. Also, coronary artery calcium (CAC) score and late gadolinium enhancement (LGE) imaging procedures may be used for detailed analyzes of coronary and cardiomyocyte function. Intra- and extracellular fluid compartment evaluations will be valuable to determine dehydration level for further studies.

Conflict of Interest: Authors declare no conflict of interest.

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Figures

Figure-1. Mean HR values for every 10 minutes during 60 minutes running test. *: Significantly higher than average first 10-min value.

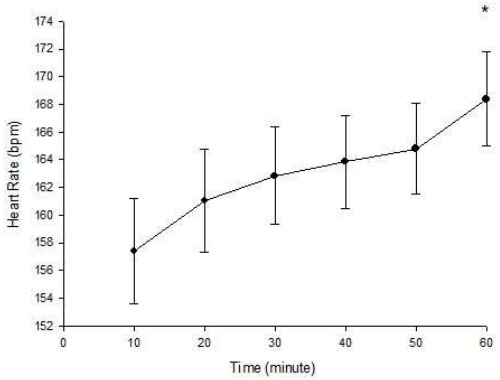


Figure-2. A- Total CK levels of the runners. *: represents a significant difference from pre-exercise, $p<0.001$, ε: represents a significant difference from post-exercise 24th hour, $p<0.05$. Dashed line represents cut-off value. B- Plasma CK-MB levels of the runners. *: represents a significant difference from pre-exercise, $p<0.001$, δ: represents a significant difference from post-exercise 4th hour, $p<0.05$, ε: represents a significant difference from post-exercise 24th hour, $p<0.05$. Dashed line represents cut-off value. C- CK index values of the runners. *: represents a significant difference from pre-exercise, $p<0.05$, #: represents a significant difference from post-exercise, $p<0.05$. Dashed line represents cut-off value.

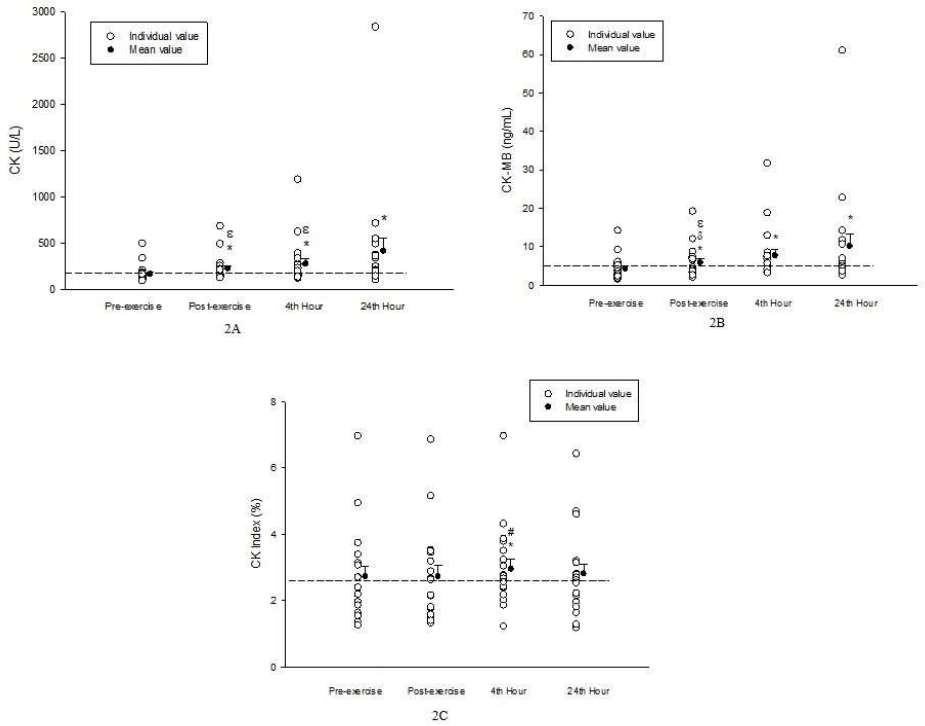
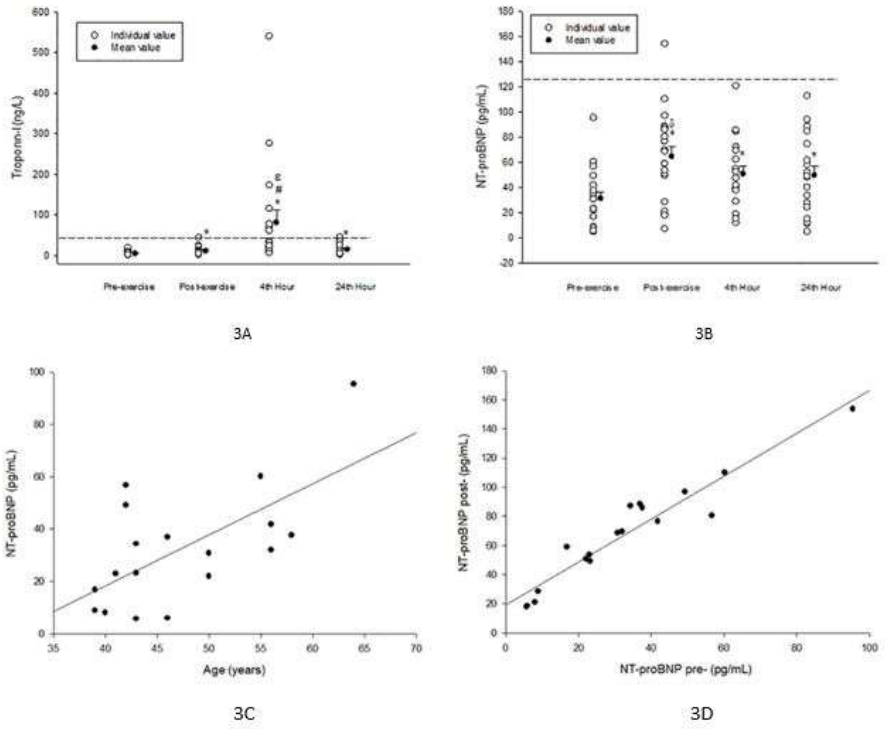


Figure-3. A- Troponin-I levels of the runners. *: represents a significant difference from pre-exercise, $p<0.001$, #: represents a significant difference from post-exercise, $p<0.001$, ε: represents a significant difference from post-exercise 24th hour, $p<0.001$. Dashed line represents cut-off value. B- NT-proBNP levels of the runners. *: represents a significant difference from pre-exercise, $p<0.01$, δ: represents a significant difference from post-exercise 4th hour, $p<0.01$. Dashed line represents cut-off value. C- The correlation between the age of the runners and the pre-exercise NT-proBNP values. D- The correlation between the initial and post-race NT-proBNP values.



Tables

	Mean±SE	Minimum - Maximum Value
Age (year)	47.4±1.7	39-64
Height (cm)	171.0±1.3	162-183
Weight (kg)	73.3±1.7	63.5–90.7
Body mass index (kg/m^2)	25.0±0.4	21.8–28.4
Fat percent (%)	18.0±0.8	11.8–27.4
Muscle percent (%)	40.4±0.5	35.8–44.5
Muscle mass (kg)	29.6±0.8	25.4–37.8

Table-1. The physical characteristics of the participants. Values presented as mean±standard error.

	Mean±SE
Resting HR (beats/minute)	66.1±2.7
Maximum HR (beats/minute)	169.2±2.8
$\dot{V}O_{2\,max}$(ml/kg/minute)	47.1±1.2
$\dot{V}O_{2\,max}$ (L/minute)	3.4±0.1
Maximum Running Speed (km/h)	16.1±0.4
AT HR (beats/minute)	136.0±2.8
AT $\dot{V}O_2$ (ml/kg/minute)	34.9±1.2
AT $\dot{V}O_2$ (L/minute)	2.6±0.1
AT Running Speed (km/h)	10.2±0.4
AT $\dot{V}O_2/\dot{V}O_{2max}$ (%)	74.3±2.2

Table-2. The CPET data (maximal and anaerobic threshold values) of the runners.

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	Pre-exercise	Post-exercise	Post-exercise 4th hour	Post-exercise 24th hour
Red Blood Cell (10^6/μL)	5.1±0.1	5.3±0.1$^{*\,\delta\,\varepsilon}$	4.9±0.1*	4.9±0.1*
White Blood Cell (10^3/μL)	6.3±0.3	8.7±0.4$^{*\,\varepsilon}$	10.4±0.7$^{*\,\varepsilon}$	6.4±0.4
Hemoglobin (g/dL)	14.8±0.2	15.3±0.2$^{*\,\delta\,\varepsilon}$	14.2±0.2*	14.3±0.3*
Hematocrit (%)	43.4±0.5	45.0±0.6$^{*\,\delta\,\varepsilon}$	41.4±0.7*	42.0±0.7*
Blood volume (%)	100.0±0.0	96.4±2.9$^{*\,\delta\,\varepsilon}$	104.3±3.6*	103.2±4.2*
Plasma volume (%)	56.6±2.3	53.1±3.6$^{*\,\delta\,\varepsilon}$	61.2±4.5*	59.9±5.1*

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555 Table-3. Venous blood sample parameters of the runners. *: represents a significant difference from pre-exercise,
556 $p<0.05$, δ: represents a significant difference from post-exercise 4th hour, $p<0.05$, ε: represents a significant difference
557 from post-exercise 24th hour, $p<0.05$.