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ORIGINAL ARTICLE

Muscle damage and inflammatory status biomarkers after a 3-stage trail running race

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ABSTRACT

BACKGROUND: Participants in ultramarathon and multi-stage races are continuously increasing. A detailed knowledge of the time-course of the restoration in muscular, cardiac, and inflammatory biomarkers after a multi-stage race may help the design of training schedules focused to avoid adverse outcomes of repetitive high-intensity endurance exercise and athlete exhaustion. Thus, the aim of the study was to evaluate blood parameters and serum biomarkers associated to muscle damage and inflammation in athletes participating in a 3-stage competition.

METHODS: Ten runners concluded the race "Magraid" consisting of 3 stages of 22, 48 and 20 km. Before (PRE), immediately after the end of the third stage (POST) and five days after the last stage (R5d), we collected blood samples.

RESULTS: Among others, at POST mean white blood cell (+57±42%; P=0.006), blood urea nitrogen (+68±39%; P<0.001), creatinine (+17±12%, P=0.005), alanine aminotransferase (ALT, +104±69%; P=0.002), lactate dehydrogenase (LDH, +116±64%; P<0.001), creatine kinase (CK, +2044±1433%; P=0.011), CK-MBm (+1544±1007%; P=0.004), cardiac troponin I (cTnI, +85±129%; P=0.015), c-reactive protein (hsCRP, +2137±1660%; P=0.015) were higher than PRE. At R5d, ALT (+72±53%; P=0.010), LDH (+32±25%; P=0.006) and hsCRP (+252±234%; P=0.021) were still different compared with PRE.

CONCLUSIONS: A 3-stage trail running race induces an inflammatory status and muscle damage and functional consequences on some physiological systems that may not be completely recovered within a short period.

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KEY WORDS: Myoglobin; Creatine kinase; Troponin; C-reactive protein; Inflammation.

Participants in ultramarathon and multi-stage races are continuously increasing[1] and the literature related to this type of events is increasing as well.[2] Consequently, several aspects of ultra-running were analyzed in the latest years (particularly, energetics, biomechanics, and fatigue).[3, 4] Some studies performed during ultra-endurance events showed that muscular damage and inflammatory biomarkers, such as myoglobin and high sensitivity C-reactive protein (hsCRP) remain elevated for days after the race,[5-7] possibly reflecting incomplete muscle recovery. In the general population, several evidences indicated that high levels of hsCRP have been related to endothelial damage, venous thrombosis, atherosclerosis, cardiovascular diseases (CVD) and several metabolic disorders.[8] Some effects of strenuous exercise are comparable to acute responses that are commonly related to injury or pathology,[9] but these are generally transient.[10] For example, an endurance running race provokes hemolysis[11] that is caused by the continuous foot strikes[12] that occurs during running, particularly during prolonged events.[13, 14] During high-intensity level running and mostly in mountain running, the eccentric work done for breaking the fall of the center of mass can lead to remarkable muscle damage.[6] Indeed, authors reported large effects on blood markers of muscle damage and inflammation after a mountain ultramarathon.[6] Notably, high levels of creatine kinase (CK) concentrations were comparable to those measured in patients undergoing severe rhabdomyolysis.[15]

The study of multi-stage races allows analyzing the recovery capability of athletes in tolerating repeated exhaustive efforts. For this type of race much information

is available in the literature, but few scientific articles examined the skeletal and cardiac muscle tissue trauma following repetitive endurance running races. High-demanding repeated efforts might cause a persistent systemic inflammatory status, which in turn may be associated with increased risk of CVD, immune dysfunction, overtraining or underperformance syndrome.[8] Thus, a detailed knowledge of the time-course of the restoration in muscular, cardiac, and inflammatory biomarkers after a multi-stage race may help the design of training schedules focused to avoid adverse outcomes of repetitive high-intensity endurance exercise and athlete exhaustion. Thus, before and at the end of a 3-stage trail running race and after five days of recovery, we evaluated red and white blood cells, and several serum biomarkers associated to muscle damage and inflammation in amateur athletes.

Materials and methods

Experimental protocol

We analyzed the data collected during the trail running competition "Magraid", performed in the North-East of Italy. This race consisted of three consecutive stages of 22, 48 and 20 km, respectively (www.magraid.it). In the week preceding the race (PRE) and five days after the last stage (R5d), participants visited the laboratory to perform a graded exercise test on a treadmill (Saturn, HP Cosmos, Germany) to evaluate their maximal oxygen uptake ($\dot{V}O2max$). Moreover, at PRE, immediately after the end of the third stage (POST) and at R5d, we collected anthropometrics and blood samples.

Subjects

We enrolled twelve healthy Caucasian male runners (age range 26-59 years) who participated in the race "Magraid" (Table I). The participants were recruited among experienced ultra-endurance runners and all of them reported to have completed a race longer than 100 km at least once in life. Also, they reported to perform on average 6.6±2.7 h/week of running. The experimental protocol was approved by the Ethics Committee of the local University, and it was conducted according to the Declaration of Helsinki. Two subjects quit the race after the first stage due to an excess of fatigue probably caused by a lack of training, and they were excluded from the statistical analysis. None of the athletes used anti-inflammatory drugs from PRE to R5d.

TABLE I.—*Anthropometric characteristics, body composition and physiological characteristics of subjects (N.=10), before (PRE) and 5 days after the recovery period (R5d).*

Parameters	PRE	R5d	P
Age (years)	38.2±12.4	-	
Stature (m)	1.76±0.07	-	
Body mass (kg)	74.5±7.3	74.6±7.5	0.780
BMI (kg/m^2)	24.1±1.6	24.2±1.6	0.534
FFM (kg)	62.3±5.2	62.7±5.5	0.087
FM (kg)	12.1±4.0	11.9±3.8	0.343
FM (%)	16.1±4.8	15.7±4.6	0.257
$\dot{V}O_2$max (mL/min)	4018±526	3750±409	0.102
$\dot{V}O_2$max (mLO_2/kg/min)	54.1±6.8	49.2±3.9	0.126
HR_{max} (bpm)	170±10	165±9	0.008
v_{max} (km/h)	16.3±1.4	16.5±1.8	0.095

All values are mean±standard deviation (SD).
BMI: Body Mass Index; FFM: fat-free mass; FM: fat mass; $\dot{V}O_2$: oxygen uptake; HR: heart rate; *v*: velocity; P: Significance by paired *t*-test.

Environmental conditions

The geologic texture of the terrain is an unusual soil in respect the vast majority of ultra-endurance competitions; it is characterized by a gravel (locally named "Magredi") from the braided river Cellina-Meduna. The first stage began at 6 p.m. with temperature and relative humidity of 31 °C and 41%. The second and third stages began at 10 a.m. with temperature and relative humidity of 17 °C and 18°C and 76% and 69%, respectively.

Anthropometrics

Body mass (BM, kg) was measured to the nearest 0.1 kg with a manual weighing scale (Seca 709, Hamburg, Germany) before and after each stage, stature was measured to the nearest 0.001 m on a standardized wall-mounted height board. Then, Body Mass Index (BMI, kg/m^2) was calculated. Body composition at PRE and at R5d was measured by bioelectrical impedance (BIA 101, Akern, Italy) by using the software provided by the manufacturer (Bodygram, 1.31). Throughout the competition, GPS coordinates and heart rate were continuously recorded by using a Garmin Forerunner 305 GPS (Garmin, Kansas City, USA).

Graded test

Before each test the volume and gas analyzers of the metabolic unit (Quark-b^2, Cosmed, Italy) were calibrated following the manufacturer's instructions. After 5 min of rest, subjects started to run at 10 km/h for 5 min (on a slope of 1%); the speed was then increased by 0.7 km/h every minute until volitional exhaustion. To calculate $\dot{V}O_2$max, we averaged the $\dot{V}O_2$ during the last minute of the test. Dur-

ing the tests, ventilatory and gas exchange responses were measured continuously, and heart rate (HR) was measured with a dedicated device (Polar, Finland).

Blood sampling

At PRE, POST and R5d, blood samples were drawn from seated subjects as previously described.[16, 17] An EDTA anticoagulated sample for full blood count and a serum sample for biochemistry were collected in Vacutainers® tubes (Belliver Industrial Estate, Plymouth, UK). Storage of aliquoted serum samples was performed at -80 °C until analysis.[18]

Evaluation of hemogram parameters was performed within 4 hours from collection of whole blood by an automatic electronic counter (Sysmex XE-2100, Sysmex America Inc., Illinois, USA).

Serum hsCRP was measured using a highly sensitive immunoturbidimetric method (Olympus), detection limit of the assay was ≤0.08 mg/L. The intra- and inter-assay coefficients of variation (CVs)% were 5.7%, and 5.8% at 0.23 mg/L, respectively, and 0.92% and 1.94% at 9.70 mg/L, respectively.

Serum CK was measured using a UV kinetic method (Olympus Life and Material Science Europe GmbH, Hamburg, Germany) by use of Olympus instrument AU2700. Detection limit of the assay was 10 IU/L. The intra- and inter-assay coefficients of variation (CVs)% were 1.3%, and 4.6% at 108 IU/L, respectively.

Myoglobin, CK-MBm and Troponin I (TnI) concentrations were measured by a quantitative immunoassay with chemiluminescent detection (Access2 Immunoassay Systems, Beckman Coulter, Inc., Fullerton, CA, USA). Myoglobin assay had detection limit 0.1 µg/L, intra CV 3.4% and inter CV 3.9% imprecision (at 29.1 µg/L). The MB isoenzyme of CK (CK-MB) was measured by a mass method (CK-MBm), assay had detection limit 0.1 µg/L, intra CV 2.6% and inter CV 3.1% imprecision (at 7.35 µg/L). A value of the ratio of CK-MBm (µg/L) over CK (IU/L) >3 was considered attesting a cardiac damage release of the CK isoenzyme.[19] Cardiac TnI (cTnI) assay had detection limit 0.01 µg/L, intra CV 2.6% and inter CV 2.8% imprecision (at 3.80 µg/L). The cTnI cut-off for acute myocardial infarction, determined as 99th percentile in the healthy population, was 0.04 µg/L.

Serum concentrations of creatinine were evaluated on modular analyzer by compensated alkaline picrate assay (Roche Diagnostics).

Serum concentrations of total bilirubin, triglycerides, albumin, aspartate aminotransferase (AST), alanine aminotransferase (ALT), blood urea nitrogen (BUN), creatinine, and glucose were measured on modular analyzer (Roche Diagnostics, Mannheim, Germany) by appropriate reagents (Roche Diagnostics) (Cauci *et al*. 2008).[18]

Statistical analysis

Data reported are referred only to the ten subjects who completed the entire race. We performed the statistical analysis using GraphPad Prism 8.3.1. We compared the parameters measured at PRE *vs*. R5d with the paired t-test, two tailed. Then, we analyzed the data collected at PRE, POST and R5d with one-way ANOVA repeated measures with the Geisser-Greenhouse correction. Later, we used Bonferroni's multiple comparisons test (two-tailed) for detecting specific differences (PRE *vs*. POST; PRE *vs*. R5d; POST *vs*. R5d). P values <0.05 were considered significant. We investigated the differences in body mass with two-way ANOVA repeated measures with two factors: stage (stage 1, stage 2, stage 3) and time (PRE and POST). Then, we used Bonferroni's multiple comparisons test for detecting specific differences (PRE *vs*. POST for each stage). We presented the results as mean±standard deviation (SD). We calculated the changes of parameter values POST and R5d *versus* PRE for each participant and we reported mean±SD variation in %.

Results

Mean cumulative running time was 9:25:50±2:12:00 hours, mean speed was 9.8±2.1 km/h and mean % HRmax was 83±4% (corresponding to 74±7% of $\dot{V}O_2$max). When considering the overall mean time of performance, it turned out that HR was maintained between 80% and 89% of its maximum for about 45% of the total race duration; it was lower than 69% and between 70% and 79% of HRmax for 15% and 20% of the total, respectively. Finally, HR was >90% HRmax for about 19% of the race duration. Body mass decreased during the stage 1 (-2.5±1.5%, P<0.001) and stage 2 (-2.0±0.9%, P<0.001) whereas it remained unchanged during stage 3 (0.0±1.2%, P>0.05).

Race and recovery effects on whole blood parameters

Values of the blood parameters measured in athletes at PRE, POST and R5d are illustrated in Table II. Several biomarkers differed significantly between PRE and POST. Mean corpuscular volume (MCV) decreased by -1.1±0.8% (P=0.006), mean corpuscular hemoglobin concentration (MCHC) increased by +1.5±1.5% (P=0.025), platelets (PLT) increased by +13.2±14.8% (P=0.047). White blood

TABLE II.—*Blood parameters determined before the race (pre-run day 1, PRE), after the end the race (postrun day 3, POST) and at the fifth day of recovery (R5d).*

N.=10	PRE	POST	R5d	P
Hemoglobin (g/dL)	14.6±1.1	14.8±1.0	14.9±0.9	0.541
Red blood cell count (1000000/microL)	4.8±0.3	4.9±0.3	4.9±0.3	0.436
HCT (%)	43.3±3.2	43.1±2.8	43.7±2.3	0.561
MCV (fL)	88.9±3.6	87.9±4.0*	88.1±3.7*	0.023
MCH (pg)	30.0±1.6	30.2±1.4	30.0±1.4	0.184
MCHC (%)	33.8±0.6	34.3±0.7*	34.0±0.8	0.024
Reticulocytes (%)	7.4±2.5	6.9±1.6	7.9±2.1	0.068
Platelet count (1000/microL)	227±57	252±47*	231±55	0.013
White blood cell count (1000/microL)	5.7±1.4	8.5±1.9*	5.1±1.2#	<0.001
Neutrophils (1000/microL)	3.3±1.1	6.2±1.8*	2.7±0.8#	<0.001
Monophils (1000/microL)	0.4±0.1	0.6±0.3*	0.5±0.1	0.018
Lymphocyte (1000/microL)	1.8±0.5	1.6±0.6	1.7±0.4	0.248
Eosinophils (1000/microL)	0.1±0.1	0.1±0.1	0.1±0.1#	0.013
Basophils (1000/microL)	0.02±0.02	0.02±0.02	0.04±0.02	0.298

All values are mean±standard deviation.
HCT: hematocrit; MCV: mean corpuscular volume; MCH: mean corpuscular hemoglobin; MCHC: mean corpuscular hemoglobin concentration; P: significance calculated with one-way ANOVA.
*Significant different from PRE; #significant different from POST.

cell (WBC) increased by +57±43% (P=0.006) and specifically neutrophils increased by +105±86% (P=0.0002) and monophils by +56± 57% (P=0.007). However, most of the parameters returned to PRE values at R5d, only MCV remained marginally different from PRE (-0.9±1.0%, P=0.045).

Many of biochemical blood parameters showed significant differences (Table III). The following parameters increased at POST: BUN (+68±39%, P<0.001), creatinine (+17±12%, P=0.005), AST (+400±286%, P=0.003), ALT (+104±69%, P=0.002), total bilirubin (+84±53%, P<0.001), uric acid (+28±17%, P=0.001), lactate dehydrogenase (LDH, +116±64%, P<0.001), CK (+2044±1433%, P=0.011), CK-MBm (+1544±1007%, P=0.004) (Figure 1A) myoglobin (+3369±2643%, P=0.001), cTnI (+85±129%, P=0.015) and hsCRP (+2137±1660%, P=0.015) (Figure 1B).

At R5d, ALT (+72±53%, P=0.010), LDH (+32±25%, P=0.006) and hsCRP (+252±234%, P=0.021) were still different compared with PRE. Other parameters returned to the PRE-values.

TABLE III.—*Blood biochemistry determined before the race (prerun day 1, PRE), after the end the race (postrun day 3, POST) and 5 days after the recovery period (R5d). Also, we reported the frequency of specified levels of myoglobin, CK-MBm and hsCRP.*

N.=10	PRE	POST	R5d	P
BUN (mg/dL)	18.2±3.8	29.9±5.28*	17.9±3.14#	<0.001
Creatinine (mg/dL)	1.1±0.1	1.32±0.21*	1.06±0.12#	0.001
BUN/creatinine	16.3±3.1	22.87±3.80*	16.9±2.43#	<0.001
AST (UI/L)	30.1±12.1	139.9±71.5*	45.0±21.7#	<0.001
ALT (UI/L)	21.6±5.7	42.8±14.74*	36.7±13.8*	<0.001
Total bilirubin (mg/dL)	0.7±0.2	1.31±0.4*	0.61±0.22#	<0.001
Total protein (g/dL)	76.7±3.4	78.65±6.58	78.12±3.40	0.403
Albumin (g/dL)	44.8±2.2	47.1±4.30	45.81±3.99	0.156
Triglycerides (mg/dL)	114.5±40.4	77.45±10.76*	135.1±48.0#	0.007
Uric acid (mg/dL)	5.6±0.9	7.17±1.01*	5.35±1.08#	<0.001
Lactate dehydrogenase (UI/L)	403.4±57.8	862.5±230.5*	530.8±101.3*#	<0.001
Myoglobin (ng/mL)	28.2±12.3	762.7±409.5*	36.10±11.3#	<0.001
Myoglobin >100 µg/L (N. [%])	0 (0%)	10 (100%)	10 (0%)	
CK (UI/L)	211.7±164.95	3637.6±2754.8*	301±186.9#	0.003
CK-MBm (µg/L)	4.42±3.96	56.50±35.99*	5.59±3.20#	0.001
CK-MBm >10 µg/L (N. [%])	1 (10%)	10 (100%)	0 (0%)	
CK-MBm/CK	2.1±0.6	1.775±0.665	1.996±0.642	0.078
Cardiac troponin I (ng/mL)	0.01±0.00	0.02±0.0*	0.01±0.01#	0.015
hsCRP (mg/L)	0.6±0.5	11.4±9.6*	1.8±1.3*#	0.005
hsCRP<0.5 mg/L (N. [%])	5 (50%)	0 (0%)	0 (0%)	
hsCRP 0.5≤1.0 mg/L (N. [%])	3 (30%)	0 (0%)	3 (30%)	
hsCRP 1.0≤3.0 mg/L (N. [%])	2 (20%)	0 (0%)	6 (60%)	
hsCRP 3.0≤10.0 mg/L (N. [%])	0 (0%)	5 (50%)	1 (10%)	
hsCRP≥3.0 mg/L (N. [%])	0 (0%)	10 (100%)	1 (10%)	
hsCRP≥5.0 mg/L (N. [%])	0 (0%)	6 (60%)	0 (0%)	
hsCRP≥10.0 mg/L (N. [%])	0 (0%)	5 (50%)	0 (0%)	

All values are mean±standard deviation.
BUN: blood urea nitrogen; AST: aspartate aminotransferase; ALT: alanine aminotransferase; CK: creatine kinase; P: significance calculated with one-way ANOVA.
*Significant different from PRE; #significant different from POST.

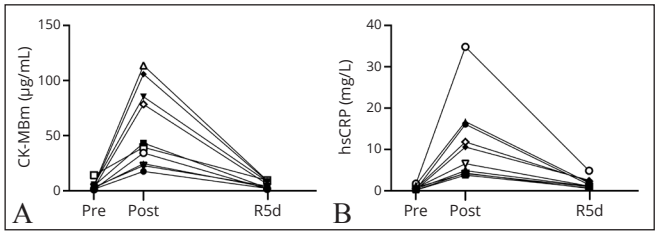

Figure 1.—A) Single participant's values of CK-MBm; and B) hsCRP at PRE, POST and R5d.

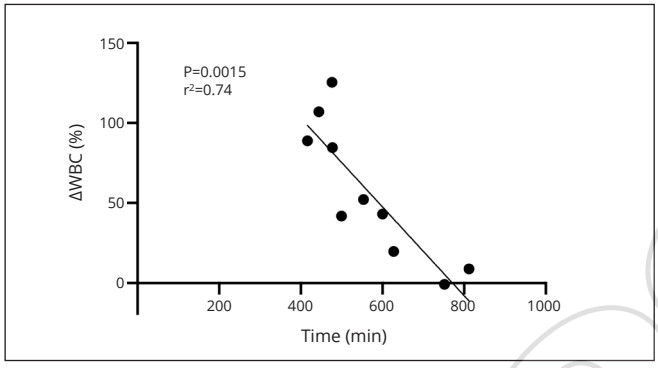

Figure 2.—Relationships between the total race time and the changes in WBC (in % POST *vs*. PRE). Fastest athletes had bigger increase in WBC.

Relationships between performance and blood parameters

Figure 2 shows that there was a significant relationship between race time and changes in WBC between PRE and POST (P=0.0015, R^2=0.74). We found no relationships between other parameters and race time.

Discussion

The main results of the present study show that a 3-stage trail running race for a total of 90 km induces: 1) a remarkable increase of inflammatory status and skeletal/cardiac muscle blood biomarkers of injury; and 2) five days of light training or rest post-race do not allow a complete recovery.

Our results confirmed that prolonged physical activity leads to a systemic inflammatory status. Indeed, in our study, inflammatory biomarkers were exceptionally elevated after three consecutive days of racing. Specifically, mean neutrophils and hsCRP were ~2-fold and ~17-fold higher at POST. Of note, at POST 50% of athletes showed hsCRP>10 mg/L indicating a remarkable inflammatory status, and 100% of them showed hsCRP>3 mg/L indicating overt inflammation.[18, 20] Although the effects of running on blood parameters are extensively reported[5, 7, 21] there are only a few papers regarding repeated effort such as a multi-stage running race.[22] We observed a 1.5-fold increase of WBC and a ~2-fold increase of neutrophils after the race. It is known that prolonged running performance affects the concentration of total white cell count, which documents the existence of an inflammatory process. The increase in WBC is considered induced by muscle damage but also by the increase of cortisol.[23] Our study confirmed that neutrophils shows the biggest changes among WBC as described previously.[23]

In parallel with an increased inflammation, several blood markers testified the muscle damage, particularly the great increase of CK (~17-fold), myoglobin (~27-fold), AST (~5-fold), ALT (~2-fold) and LDH (~2-fold) levels. To assess the cardiac damage the CK-MBm and cTnI levels are commonly used.[5] We observed CK-MBm values higher at POST (~14-fold), and all athletes had CK-MBm >10 µg/L that is considered a marker of acute myocardial infarction in non-athletes.[24] However, none of the athletes in POST had a ratio of CK-MBm/CK above 3, thus indicating the non-cardiac origin of the circulating CK-MB isoenzyme.[19] Moreover, only one athlete had cTnI above (0.05 µg/L) the healthy reference value threshold of 0.04 µg/L.

It is well-known that hsCRP increases in the acute phase when an inflammation or infection occurs, as well as after a heart attack. The elevated values of CK-MBm, cTnI and hsCRP that we observed, are similar to the trend following a heart attack.[24] However, none of the participants had evidence of cardiac complications after the race.

Conversely, there is evidence that physical activity is able to reduce the CVD risk.[25, 26] An intriguing hypothesis is that one of the mechanisms by which physical exercise may reduce the risk of vascular thrombosis and CVD is the reduction of resting hsCRP levels.[7] This hypothesis is of great interest for the primary prevention of the CVD.[27] However, data gathered up to now on the effects of physical activity on plasma hsCRP levels are contradictory. Intense endurance physical activity has the potential to worsen the hsCRP profile, likely as a consequence of inflammatory process following muscle damage. We found that 50% of athletes had CVD protective levels of hsCRP<0.5 mg/L before race, but all athletes had CVD risky levels (hsCRP>3 mg/L) at the end of the race. Notably, no athlete had protective hsCRP levels five days post-race. Thus, we assessed that in endurance athletes the rate of elevated hsCRP is highly modified by the race and pre-race levels cannot be restored within five days.

In our study we observed no changes in most of RBC-related parameters (RDW, reticulocytes, HCT, Hb, MCH). This is in contrast with previous study in which linear changes (increase or decrease) were reported during a five-days stage race[22] or after four days of a continuous event (no-stop running) of total 1600 km in which athletes covered on average ~96 km/day.[28] Indeed, these studies reported decrease in Hb and HCT and increase in PLT. Particularly, changes in HCT may be affected by the hydration status of the subjects. In our study, the low change in body mass (~2%) between PRE and POST each stage (an indirect marker of hydration status) did not affect the hemoconcentration. Further, we observed only a slight increase in PLT that agrees with other studies[29, 30] and it may be linked to cardiovascular complications.[8, 31]

Also, we observed an increase in bilirubin that is usually due to hemolysis in competing athletes, which may be probably caused by continued impact with the ground.[12, 32] The repeated efforts required by a multi-stage race may accentuate the hemolysis. Indeed, we reported an increase of ~84% of bilirubin which is slightly higher than the ~70% reported by others.[28] Unfortunately, we have no data about haptoglobin to confirm this hypothesis. After three days of race the kidney function resulted partially impaired as the BUN and creatinine resulted higher than normal. This condition is usually reported after renal failure which produces a ratio BUN: creatinine higher than 10:1.[33] To note that the athletes that we enrolled presented a ratio higher than the normal values (~16) at PRE and it increased to ~21 after the third stage. Also, the uric acid increase is likely associated to kidney function alterations. This may indicate that continuous running efforts should be followed by long period of active and passive recovery to restore the functionality of this system.

The differences of our study compared with others may be attributed to the different race conditions (distance, environment, temperature). Also, the fitness of the athletes may affect the results. We found an inverse relationship between the increase in WBC and the race time and this suggest that fastest athletes can suffer higher muscle damage, probably because of the faster speed maintained during the race and greater ground reaction forces that act on the lower limbs' musculature. This agrees with a study that reported changes in many blood parameters with differences between the fastest and slowest athletes.[22]

In general, there is poor attention to the health status of asymptomatic endurance athletes performing physical activity at non-professional level. Despite recreational athletes account for a much larger proportion of the endurance athletes, the ones that we enrolled are not invited to check and take care of their health status like elite athletes. These results may help clinicians to correctly interpret serum biomarkers values observed in patients who perform ultra-endurance activity. Further, our data can support a rational approach to post-race supplementation treatments.[34]

Conclusions

A 3-stage trail running race induce an inflammatory status and muscle damage and functional consequences on some physiological systems that may not be completely recovered with a short period. We suggest maintaining under control some parameters to avoid pathological complications in non-professional sport population.

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