

• CLINICAL RESEARCH •

# Effects of 24 h ultra-marathon on biochemical and hematological parameters

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## Abstract

**AIM:** To analyze detailed changes in hematology and biochemistry tests parameters before and after a long-distance race in ultramarathon runners.

**METHODS:** Blood samples of 11 participants were obtained for standard analysis before, immediately after, two days after and nine days after the 2002 International Ultramarathon 24 h Race and the International Association of Ultrarunners (IAU) Asia 24 h Championship.

**RESULTS:** Total bilirubin (BIL-T), direct bilirubin (BIL-D), alkaline phosphatase (ALP), aspartate aminotransferase (AST), alanine aminotransferase (ALT) and lactate dehydrogenase (LDH) increased statistically significantly ( $P<0.05$ ) the race. Significant declines ( $P<0.05$ ) in red blood cell (RBC), hemoglobin (Hb) and hematocrit (Hct) were detected two days and nine days after the race. 2 d after the race, total protein (TP), concentration of albumin and globulin decreased significantly. While BIL, BIL-D and ALP recovered to their original levels. High-density lipoprotein cholesterol (HDL-C) remained unchanged immediately after the race, but it was significantly decreased on the second and ninth days after the race.

**CONCLUSION:** Ultra-marathon running is associated with a wide range of significant changes in hematological parameters, several of which are injury related. To provide appropriate health care and intervention, the man who receives athletes on high frequent training program high intensity training programs must monitor their liver and gallbladder function.

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## INTRODUCTION

Numerous reports have been published on the effects on the body of endurance sports such as the 5 000 m, the 10 000 m, the marathon, cross-country running, rowing and cycling<sup>[1-14]</sup>. In recent years, more athletes have become involved in ultra-endurance races, such as the iron-man triathlon, the 100 km race and the 24 h marathon, and a few investigations have addressed the related hematological and biochemical changes. Strenuous physical activities are becoming increasingly popular around the world, and this work may benefit participants in future competitions.

Some of these previous studies have involved staged races with long rest periods; while others made comparisons before and after competition. This study examines, athletes who have completed a long-distance run lasting 24 h, and detailing the related hematological and biochemical changes before, immediately after, two days after and nine days after the race.

## MATERIALS AND METHODS

### Anthropometric data

A 24 h ultra-marathon was held at Soo-Chow University on March 2, 2002. The runners ran around a 400 m oval track for 24 h, covering a distance of at least 100 km. The runners changed direction every 4 h. The temperature during the 24 h race ranged from 19.0 to 26.8 °C and the relative humidity ranged from 63 to 91%. The runners were permitted to rest and to ingest water and food freely. Ten males (10/36) and one female (1/1) participated in this study, having previously given their informed consent. Table 1 lists the anthropometric data.

**Table 1** Anthropometric data of the study population ( $n = 11$ )

	Range	Mean	Standard Deviation
Age (yr)	26.00-55.00	45.10	±2.64
Height (cm)	155.00-177.00	166.80	±6.23
Body weight (kg)	47.00-69.30	60.60	±9.69
Distance completed (km)	106.70-194.40	158.60	±26.78
BMI(body mass index)	19.56-22.12	21.79	±0.24

### Parameter of blood tests

20 mL blood samples were obtained from the antecubital vein 24 h before the race, immediately after the race, two days after the race and nine days after the race. The blood was analyzed in 1 h using an ABBOTT CELL DYN 3 000 autoanalyser (Abbott Diagnostics, Mountain View, CA., USA) and HITACHI 7150 autoanalyser (Hitachi High Technologies, Tokyo, Japan).

Statistical significance of paired differences in means and standard deviations of the related hematological and biochemical changes among pre-race, immediately post-race, two days post-race, and nine days post-race values were calculated using by one-way ANOVA analysis. The level of significance was set at  $P<0.05$ .

## RESULTS

### Hb. Hct. platelet data

The pre-race red cell count, Hb and Hct levels were not significantly

immediately after the race, but were significantly reduced two and nine days after the race, being lowest at two days after the race. The mean cell volume was not significantly changed immediately after the race or on day two post-race, but was significantly increased by day nine post-race. Mean cell Hb concentration was significantly lower on day two than before the race, but had recovered on day nine.

Mean cell Hb and red cell distribution width remained unchanged at all times. The platelet concentration immediately after the race and on day nine following the race was increased significantly compared to pre-race values, but had decreased significantly on day two. (Table 2).

#### Total WBC and differential count

The white blood cell count was significantly increased at the end of the race and remained high until day nine. Moreover, the number of neutrophils was increased at the end of race but recovered two days later. Furthermore, the lymphocytes, eosinophils and basophils were decreased immediately after the race but recovered two days later. Finally, the number of monocytes was increased immediately after the race and on day two post-race, but returned to the pre-race level by day nine post-race. (Table 3).

#### Ferritin, TIBC

The ferritin level, total iron binding capacity (TIBC) and transferring saturation rose significantly immediately post-race along with ferritin level, and remained at the end higher on day two and nine. All of the parameters remained normal

mean values. (Table 4).

#### Liver function tests

The BIL-T and BIL-D concentrations were significantly raised immediately after the race and normalized two days later. TP, albumin and globulin concentrations were unchanged immediately after the race but were significantly reduced on day two, recovering gradually after day nine, though TP and albumin remained below pre-race levels.

ALP, AST and ALT had increased significantly by the end of the race. ALP returned to its pre-race level after day two. Moreover, AST declined by day two and resumed its pre-race level by day nine. Furthermore, ALT continued to rise until day two and had recovered by day nine. Gamma glutamyl transferase ( $\gamma$ -GT) remained unchanged until the end of race and beyond the end of the event. Finally, LDH was significantly raised by the end of the race and was decreased on day two post-race, but remained above their pre-race level on day nine. (Table 5).

#### Lipid metabolism

Triglyceride (TG), cholesterol/low-density lipoprotein cholesterol (CHO/LDL-C) ratio and LDL-C were lower immediately after the race had finished. TG level and CHO/LDL-C ratio recovered by day two post-race, while LDL-C recovered by day nine. Cholesterol was not significantly changed at the end of the race, but was significantly lower on day two.

HDL-C was highest immediately after the race but had reduced by day two and nine post-race. (Table 6).

**Table 2** Changes in Hb, Hct, red cell parameters and platelet count before and after the race

	Pre-race	0 h post-race	2 d post-race	9 d post-race
Red cell count ( $\times 10^{12}/L$ )	4.71 $\pm$ 0.25 <sup>ce</sup>	4.71 $\pm$ 0.45	4.07 $\pm$ 0.27	4.42 $\pm$ 0.21
Hb (g/dL)	14.63 $\pm$ 0.91 <sup>ce</sup>	14.58 $\pm$ 1.17	12.52 $\pm$ 0.86	13.81 $\pm$ 0.69
Hct (%)	42.34 $\pm$ 2.73 <sup>ce</sup>	42.37 $\pm$ 3.82	37.33 $\pm$ 3.15	40.27 $\pm$ 1.84
Mean cell volume (fl)	89.91 $\pm$ 3.11 <sup>e</sup>	90.05 $\pm$ 3.37	90.29 $\pm$ 3.50	91.15 $\pm$ 3.19
Mean cell Hb (pg)	31.09 $\pm$ 1.23	31.02 $\pm$ 1.44	30.90 $\pm$ 1.29	31.22 $\pm$ 1.34
Mean cell Hb				
Concentration (g/dL)	34.59 $\pm$ 0.45 <sup>e</sup>	34.44 $\pm$ 0.60	34.24 $\pm$ 0.47	34.25 $\pm$ 0.64
Red cell distribution width (%)	12.84 $\pm$ 0.60	12.94 $\pm$ 0.88	12.69 $\pm$ 0.57	12.80 $\pm$ 0.65
Platelet ( $\times 10^9/L$ )	235.45 $\pm$ 47.27 <sup>ace</sup>	248.91 $\pm$ 46.95	209.82 $\pm$ 58.28	280.27 $\pm$ 67.23

<sup>a</sup> $P$ <0.05 vs statistically significant when pre-race compared with 0 h post-race. <sup>c</sup> $P$ <0.05 vs statistically significant when pre-race compared with 2 d post-race. <sup>e</sup> $P$ <0.05 vs statistically significant when pre-race compared with 9 d post-race.

**Table 3** Total and differential white cell counts before and after the race

	Pre-race	0 h post-race	2 d post-race	9 d post-race
White cell count ( $\times 10^9/L$ )	4.95 $\pm$ 1.05 <sup>ace</sup>	11.87 $\pm$ 1.46	5.83 $\pm$ 1.09	5.95 $\pm$ 1.45
Neutrophils (%)	56.02 $\pm$ 6.69 <sup>a</sup>	76.43 $\pm$ 6.28	57.66 $\pm$ 7.28	57.93 $\pm$ 9.16
Lymphocytes (%)	33.10 $\pm$ 6.94 <sup>a</sup>	14.47 $\pm$ 4.82	30.89 $\pm$ 6.42	32.15 $\pm$ 8.07
Monocytes (%)	7.83 $\pm$ 3.58	8.21 $\pm$ 2.84	9.34 $\pm$ 2.77	7.15 $\pm$ 1.91
Eosinophils ( $\times 10^9/L$ )	2.07 $\pm$ 1.01 <sup>a</sup>	0.20 $\pm$ 0.19	1.75 $\pm$ 0.76	1.80 $\pm$ 1.20
Basophils ( $\times 10^9/L$ )	0.96 $\pm$ 0.19 <sup>a</sup>	0.68 $\pm$ 0.24	0.82 $\pm$ 0.26	0.99 $\pm$ 0.28

<sup>a</sup> $P$ <0.05 vs statistically significant when pre-race compared with 0 h post-race. <sup>c</sup> $P$ <0.05 vs statistically significant when pre-race compared with 2 d post-race. <sup>e</sup> $P$ <0.05 vs statistically significant when pre-race compared with 9 d post-race.

**Table 4** Comparisons of parameters related to iron metabolism before and after the race

	Pre-race	0 h post-race	2 d post-race	9 d post-race
Ferritin ( $\mu g/L$ )	64.45 $\pm$ 27.95 <sup>ae</sup>	117.00 $\pm$ 52.66	70.18 $\pm$ 44.88	103.36 $\pm$ 42.15
TIBC ( $\mu mol/L$ )	361.00 $\pm$ 31.38 <sup>ae</sup>	372.18 $\pm$ 30.93	357.64 $\pm$ 35.43	356.36 $\pm$ 30.75
Transferrin saturation (%)	17.73 $\pm$ 8.05 <sup>ae</sup>	31.09 $\pm$ 13.32	19.27 $\pm$ 11.62	29.18 $\pm$ 11.70

<sup>a</sup> $P$ <0.05 vs statistically significant when pre-race compared with 0 h post-race. <sup>c</sup> $P$ <0.05 vs statistically significant when pre-race compared with 2 d post-race. <sup>e</sup> $P$ <0.05 vs statistically significant when pre-race compared with 9 d post-race.

**Table 5** Serum enzyme activity before and after the ultra marathon race

	Pre-race	0 h post-race	2 d post-race	9 d post-race
BIL-T (μmol/L)	11.63±2.91 <sup>a</sup>	25.65±9.75	13.68±7.70	12.14±4.10
BIL-D (μmol/L)	2.57±0.68 <sup>a</sup>	7.01±2.91	3.25±1.54	2.74±1.20
TP (g/L)	72.51±4.70 <sup>ce</sup>	72.50±6.21	66.14±3.90	67.00±4.91
Albumin (g/L)	44.82±2.83 <sup>ce</sup>	45.42±2.92	38.55±5.83	42.43±2.84
Globulin (g/L)	27.53±2.51 <sup>c</sup>	27.25±3.74	25.56±2.01	27.53±2.42
ALP (U/L)	132.85±56.50 <sup>a</sup>	160.55±33.00	131.36±34.00	134.27±34.40
AST (U/L)	37.10±19.10 <sup>ac</sup>	536.70±311.10	271.30±227.80	34.30±8.70
ALT (U/L)	35.10±13.10 <sup>ace</sup>	118.40±75.10	126.00±68.30	50.50±18.90
γ-GT (U/L)	20.18±6.23	24.18±14.30	19.18±9.00	20.91±8.00
LDH (U/L)	367.50±105.60 <sup>ace</sup>	1 420.50±598.50	1 120.30±605.10	582.70±207.90

<sup>a</sup>*P*<0.05 vs statistically significant when pre-race compared with 0 h post-race. <sup>c</sup>*P*<0.05 vs statistically significant when pre-race compared with 2 d post-race. <sup>e</sup>*P*<0.05 vs statistically significant when pre-race compared with 9 d post-race.

**Table 6** Changes in parameters related to lipid metabolism before and after the ultra marathon race

	Pre-race	0 h post-race	2 d post-race	9 d post-race
TG (mmol/L)	0.95±0.27 <sup>a</sup>	0.67±0.28	0.84±0.32	1.09±0.55
CHO (mmol/L)	4.87±1.06 <sup>c</sup>	4.63±1.09	4.13±0.57	4.51±0.50
HDL-C (mmol/L)	1.92±0.47	2.02±0.50	1.68±0.24	1.77±0.32
LDL-C (mmol/L)	2.51±0.78 <sup>ac</sup>	2.30±0.72	2.02±0.49	2.24±0.39
CHO/ HDL-C	2.50±0.48 <sup>a</sup>	2.30±0.37	2.40±0.38	2.60±0.43

<sup>a</sup>*P*<0.05 vs statistically significant when pre-race compared with 0 h post-race. <sup>c</sup>*P*<0.05 vs statistically significant when pre-race compared with 2 d post-race. <sup>e</sup>*P*<0.05 vs statistically significant when pre-race compared with 9 d post-race.

## DISCUSSION

Few studies have extensively addressed the hematological and biochemical changes in endurance runners. This investigation elucidates the effects of intensive exercise on athlete health and the findings can be used to help participants in future competitions.

Red cell count, Hb and Hct, three indicators of anemia, were normal before the race. Significant decreases was found by day two, consistent with the accelerated destruction of RBC in endurance athletes. The three indicators remained reduced between days two and nine; so-called sports anemia<sup>[15]</sup>, is not only caused by hemolysis owing to mechanical trauma but also by oxidative injuries of the red cells<sup>[16]</sup>. Under normal conditions, red cells with a mean life of 120 d are renewed at approximately 1% daily. However, this turnover rate increases following endurance training, as reflected in the participants in this study. The increased turnover rate is good for the athletes as the young red cells can carry oxygen more efficiently than the older cells.<sup>[17,18]</sup> The mean cell volume, mean cell hemoglobin and mean cell hemoglobin concentration remained normal throughout. The transient sports anemia was caused by reduced red cell numbers rather than red cell size or amount of Hb<sup>[19,20]</sup>. The change in platelets number was inconsistent with previous studies. The platelet count was higher at the end of the race and on day nine, but remained within the normal range, and no coagulopathy was detected. Further study can clarify the significance of the increase. The white cell count increased markedly race and subsequently declined. The initial increase followed from a rise in peripheral reserves and was mostly associated with neutrophils. Neutrophilia and numbers of lymphocytes were related to catecholamine, cortisol and some chemotactic factors: transient immunological dysfunction may occur under such conditions<sup>[21,22]</sup>. This study found on decrease in absolute lymphocyte count and no signs of infection in a follow-up questionnaire administered 2 wk after the race.

The concentration of serum ferritin was significantly increased immediately after the race and on day nine post-race, owing to the acute phase response of the destruction of red cells, consistent with previous reports. Total iron binding capacity and transferrin saturation were markedly increased by

the end of race, reflecting the acute release of iron.

BIL and BIL-D increased at the end of the race and normalized after day two, associated with hemolysis that follows from ultra-long running. The hemolysis was related to a decline in haptoglobin concentration and structural changes in the red cell membranes<sup>[23,24]</sup>. AST, ALT, γ-GT, LDH and ALP all increased by the end of the race, implying damage to the skeletal muscle cells and hepatic cells. Serum BIL normalized by day two as red cell turnover reduced, but AST, ALT and LDH continued to exceed pre-race levels, representing a continued release of enzymes from the muscles and liver<sup>[25-27]</sup>.

Albumin is involved in protein synthesis by the liver. Albumin reduced significantly by day two, reflecting damage to the anabolic functioning of hepatic cells. TP fell after day two, mainly owing to the decrease in albumin and had not recovered by day nine. Despite the reduced protein level, no clinical pitting edema was found. The pre-race mean AST, ALT and LDH exceeded the normal range, possibly indicating chronic damage to the liver following long-term strenuous exercise.

Most hepatic function parameters displayed no correlation with age, except for negatively correlated globulin, reflecting the lower immunological functioning of older runners following endurance exercise<sup>[28,29]</sup>. AST, ALT and LDH were positively correlated with runner performance and unrelated to BIL and ALP, implying that changes in hepatobiliary parameters resulted mainly from damage to hepatic cells. Long-term regular exercise has been recognized to contribute to reducing cholesterol<sup>[30-32]</sup>, triglyceride and LDL-C and increasing HDL-C. In this study the lipid parameters of all of the participants were in the normal ranges, supporting the beneficial effect of rhythmic aerobic exercise. However, the effects of long-term ultra-endurance activities deserve further investigation.

Most fat is stored as triglyceride in fat and muscle cells. Plasma and muscular triglyceride were consumed equally during the first stage of endurance exercise, and subsequently the free fatty acid became the major source of energy explaining the reduction in triglyceride at the end of the race<sup>[33,34]</sup>. The TG and LDL-C were significantly lower at the end of the race and on day agreeing with previous reports. Cholesterol is a major

risk factor for coronary artery disease and deserves extensive investigation. Cholesterol levels were not significantly changed at the end of the race, and were decreased by day two, but the cholesterol/HDL-C ratio, which is not affected by plasma volume, was significantly reduced by the end of the race. This phenomenon may result from the increase in HDL-C and the decrease in LDL-C.

Ultramarathon running is associated with numerous changes in hematological parameters, many of which are injury related. These changes should not be confused with indicators of disease. Increasing liver enzyme levels in runners indicated damage to liver cells, but increased BIL resulted from higher clearance rates of RBC. The liver damage was directly proportional to workload. Acute reduction of TG might result from the use of body fat as the major energy source. An ultra-long endurance run effectively reduced LDL-C for two days post-race, but did not significantly change HDL-C.

Safety guidelines, protective equipment and prevention education are crucial to reducing sports injuries. Then, preventing liver and gall bladder injuries and ensuring safe health management program are necessary for ultramarathon athletes. In summary, efforts to minimize these injuries are warranted both to ensure the long-term health of runners and to reduce medical costs. The key to management of ultramarathon runner osteoporosis involves identifying the potential risk for osteoporosis and osteoporotic fracture, followed by measures that focus on reducing modifiable risk factors through health management program<sup>[33-34]</sup>.

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