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# Circulating microRNAs after a 24-h ultramarathon run in relation to muscle damage markers in elite athletes

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## Introduction

Ultra-endurance sports are growing in popularity but can be associated with adverse health effects, such as exercise-induced muscle damage (EIMD), which can lead to exertional rhabdomyolysis. Although delayed loss of muscle function (i.e. 24-48 h) is considered to be the best indirect marker to evaluate the magnitude of EIMD, measurements within a few hours after the end of exercise may reflect a combination of muscle damage and muscle fatigue rather than muscle damage alone. Moreover, the most commonly used biomarkers of EIMD (Creatine kinase and Myoglobin) are affected by high inter-individual variability and does not necessarily reflect the magnitude of delayed loss of muscle function. MicroRNAs (miRNAs), small non-coding RNA involved in the post-transcriptional regulation of gene expression, may be useful for the accurate evaluation of the degree of EIMD. Indeed, muscle specific/enriched miRNAs (also called myomiRs) can be actively secreted by or passively leak out of injured myocytes and measured in the blood. Thus, we aimed to: 1) investigate the relevance of circulating myomiRs as biomarkers of muscle damage and 2) examine the acute response of skeletal/cardiac muscle and kidney biomarkers to the 24-h run World Championships in elite athletes.

## Methods

Athletes of the French national team (n = 11) participated in the 24-h Run World Championships. Counter-movement jump (CMJ), delayed-onset muscle damage (DOMS), creatine

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kinase (CK), myoglobin (Mb), creatinine (Cr), high-sensitivity cardiac troponin T (hs-cTnT) and muscle-specific miRNAs (myomiRs) levels were measured before (PRE), immediately after (POST), 24 and 48h after the race.

## Results

CMJ height was reduced immediately after the race ( $-84.0 \pm 25.2\%$ ,  $p < 0.001$ ) and remained low at 24h ( $-43.6 \pm 20.4\%$ ,  $p = 0.002$ ). DOMS ( $53 \pm 30$  mm,  $p < 0.05$ ), CK activity ( $53,239 \pm 63,608$  U/L,  $p < 0.001$ ) and Mb concentration ( $9,748 \pm 7,997$  ng/mL) increased immediately after the race and remained elevated 24h after ( $p < 0.01$ ). The hs-cTnT concentrations were significantly higher at POST ( $34.1, \pm 18.3$  ng/L,  $p < 0.001$ ) compared to PRE values ( $6.3 \pm 2.3$  ng/L). There were no significant differences in Cr levels or estimated glomerular filtration rate between PRE and POST, 24 h or 48, but the values were significantly lower at 24-48 h than at POST ( $p < 0.01$ ). Circulating myomiRs levels (miR-1-3p, miR-133a-3p, miR-133b, miR-208a-3p, miR-208b-3p, and miR-499a-5p) were elevated immediately after the 24-h run (fold changes: 18–124,723,  $p < 0.001$ ). The CMJ height loss at 24h was significantly ( $p < 0.05$ ) or tended to be significantly ( $p < 0.07$ ) correlated with a set of myomiRs, but not with CK ( $p = 0.23$ ) or Mb ( $p = 0.41$ ) values.

## Conclusion

All elite ultramarathon runners included in our study were diagnosed with hyperCKemia (CK  $> 10,000$  U/L) after the 24-h run World Championships. However, despite the high level of sarcoplasmic proteins into the bloodstream, the deterioration of kidney function was moderate and transient in this population. The elevation of cardiac biomarkers was likely due to exercise-related cytosolic release rather than cardiac necrosis. Importantly, the present study is the first to show that the circulating levels of muscle-specific miRNAs may reflect the muscle function loss 24h after EIMD, suggesting myomiRs may be a useful to approach the magnitude of muscle damage. MyomiRs may have a future interest in the diagnosis of muscle damage both in sport medicine and in the monitoring of physical activity.

**Keywords:** exercise, rhabdomyolysis, ultra endurance, muscle function, cardiac stress, acute kidney injury, biomarkers