**Title**:

The concentration of soluble intercellular adhesion molecule-1 (sICAM-1) in blood before and after exercise-induced muscular damage with or without the use of whole-body cryostimulation in triathlon athletes.

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**Abstract**

We present the changes in the blood concentration of soluble intercellular adhesion molecule-1 (sICAM-1) before and after exercise-induced muscular damage in triathlon athletes. We investigated whether the use of whole-body cryostimulation (3 min at -110°C) was able to induce any changes in circulating sICAM-1.

**Specifications Table**

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| --- | --- |
| Subject area | *Biology and exercise physiology* |
| More specific subject area | *Cryobiology* |
| Type of data | *Text file and figure* |
| How data was acquired | *Data was acquired after analyzing blood specimens (using ELISA kits) obtained from athletes* |
| Data format | *Analyzed* |
| Experimental factors | *No special pretreatment of samples* |
| Experimental features | 11 male endurance athletes performed twice (randomized crossover design) strenuous running leading to *exercise-induced muscle damage*, followed by passive recovery or the use of whole-body cryostimulation. Blood specimens were taken before and at different time after the end of exercise. |
| Data source location | *Paris and Poitiers, France* |
| Data accessibility | *Data is with this article* |
| Related research article | *Bieuzen F, Hausswirth C, Dugué B. Circulating soluble intercellular adhesion molecule-1 (sICAM-1) after exercise-induced muscular damage: does the use of whole-body cryostimulation influence its concentration in blood? Cryobiology 2019, in press*and *Pournot H, Bieuzen F, Louis J, Mounier R, Fillard JR, Barbiche E, Hausswirth C. Time-course of changes in inflammatory response after whole-body cryotherapy multi exposures following severe exercise. PLoS One. 2011;6(7):e22748**We had from this previous work carefully stored aliquots of plasma specimens that were analyzed for sICAM-1 in the present report.* |

**Value of the Data**

* Our data are putting to an end the current discussion whether the concentration of sICAM-1 in blood increase or decrease after exercise and cold stimulation (cryostimulation).
* *circulating sICAM-1 biovariability can be analyzed for well trained male athletes and the within- and between-subject variation coefficients concerning this analyte be calculated*
* Based on the obtained biological variation data a significant clinical change in sICAM-1 concentration can be calculated for male athletes

**Data**

It has recently been questioned whether intercellular adhesion molecule-1 (ICAM-1) and its soluble form (sICAM-1) are key components in the mechanisms involved in the exercise-induced muscular damage (EIMD) and whether whole-body cryostimulation (WBC; exposure of 2 to 4 min at -110°C or less after exercise) which lower EIMD could also impact the amount of circulating soluble ICAM-1 (Ferreira-Junior et al., 2014, Front Physiol 5, 247; Dugué, 2015, Front Physiol 6, 35). However, the discussion was very theoretical as no data on the changes of sICAM-1 due to the combine effects of EIMD and whole-body cryostimulation were available.

Interestingly, our colleagues from Paris (INSEP, France) had in their freezers well conserved plasma aliquots obtained from athletes who experienced EIMD with and without cryostimulation at the end of exhausting exercise (Pournot et al, PloS ONE 2011;6, e22748). In this previous experiment, athletes experienced EIMD that led to muscle soreness accompanied with pro-inflammatory response. The use of whole-body cryostimulation (-110°C, 3min of exposure) after the exercise enabled both a reduction in muscle soreness and in the concentration of circulating markers of inflammation (lower blood concentration in C-reactive protein and interleukin-1beta) and an increase in the concentration of anti-inflammatory circulating markers (increase in the concentration in blood interleukin-1 receptor antagonist).

Therefore, we decided to determine sICAM-1 in our well preserved specimens from the previously mentioned experiment in order to get information on how sICAM-1 may change after EIMD and cryostimulation challenge.

**Experimental Design, Materials, and Methods**

The subjects and the methods have previously been described (Pournot et al, PLoS ONE (2011); 6:e22748). Eleven well-trained runners (age 31.8 ± 2.0 years; VO2max 62.0 ± 1.2 ml.min-1.kg-1) participated in the study. All of them gave their informed consent and the study was approved by the local Ethics Committee (Île-de-France XI, France; Ref. 200978).

Study Design

The study had a crossover design. The subjects completed two simulated runs at a one-month interval on a treadmill followed in a random order by a passive recovery or with a recovery where WBC was used. The runs were organized on the same treadmill and were designed to generate fatigue.

Blood specimens were collected before and after the simulated run, after the first recovery session, and before the recovery sessions after 24 h, 48 h, 72 h, 96 h following the end of the run. Hydration/nutrition before and during each session was standardized. Between trials, low intensity training was ensured. During the whole experiment period the subjects refrained from consumption of any anti-inflammatory pills and the use of any additional methods to improve their recovery.

One week before the first simulated run, subjects were familiarized with the test scheme and preliminary tests were performed to determine maximal oxygen uptake, the first and the second ventilatory thresholds, and the maximal aerobic speed as previously described (Pournot et al, PLoS ONE (2011); 6:e22748). Moreover, the subjects were exposed individually to a one-time session of extremely low temperature (−110°C) in a cryogenic chamber (Icelab®, Zimmer MedizinSysteme, Ulm, Germany).

Simulated Run

The run was designed to mimic race conditions encountered in a trail run. The race lasted 48 minutes and was divided into 5 stages. The first stage consisted of a 6 minute run on treadmill on a flat surface (0% gradient), followed by 3 minute uphill (+10% gradient) and 3 minute downhill (−15% gradient). At 0% gradient the exercise intensity corresponded to an intensity comprised between the first and the second ventilatory thresholds of the subject; at +10% gradient the velocity corresponded to ≈80% of the maximal aerobic velocity of the subject; and at −15% gradient the velocity corresponded to the velocity observed at the first ventilator threshold during the preliminary test. Stages 2–5 consisted of 3 min at 0°, followed by 3 min uphill and 3 min downhill at the gradients and velocities previously described (Pournot et al, PLoS ONE (2011); 6:e22748).

Recovery Modalities (WBC vs. passive)

As the study had a crossover design, subjects were randomly assigned to WBC or passive recovery after the simulated run. WBC sessions were administered in a cryogenic chamber (Zimmer MedizinSysteme GmbH, Ulm, Germany) and the subjects were exposed to -110°C for 3 min. Before the exposure, subjects were instructed to dry any sweat, and clothe themselves in swimming trunks, a surgical mask, ear bands, gloves, dry socks and sabots. After the WBC session, subjects spent 10 min comfortably seated in an environment at 24°C wearing a bath robe. The control recovery was a passive recovery during which each subject was comfortably seated in an armchair for 30 min at 24°C. The subjects were not allowed to speak to anyone during the experiment.

Biochemical Analyses

Blood specimens were analyzed in a single batch at the end of the study in order to avoid inter-assay variation. Only hematological measures were performed on the day of collection. Blood specimens were collected from a superficial forearm vein using standard venipuncture techniques and EDTA tubes (Greiner Bio-one; Frickenhausen, Germany).

Blood picture - Blood specimen were analyzed for leukocyte and erythrocyte count using an automated cell counter (Cell-Dyn® Ruby™, Abbott, IL, USA).

sICAM-1 measurements

ELISA kits for the determination of sICAM-1 were purchased from R&D Systems Europe. The intra-assay imprecision (CVA) was 4.6% and 4.8% for sICAM-1 concentrations of 126 and 476 mg/L, respectively. All blood samples were analyzed in duplicate on a spectrophotometer Dynex MRXe (Magellan Biosciences, Chelmsford, MA, USA).

Statistical Analyses

Results are expressed in Figure 1 as means ± SD. Two-way analysis of variance for repeated measurements was used to analyze the changes between cold and control experiments with time and treatments (WBC versus control) as factors. We used logarithm transformation as the data distribution was not Gaussian. Post hoc multiple comparisons were made by PLSD test when appropriate to single out statistical significance. Statistical significance was set at p<0.05.

Moreover, circulating sICAM-1 biovariability can be analyzed (Fraser & Harris, *Crit Rev Clin Lab Sci* (1989); **27**:409-437) and the within- and between-subject variation coefficients concerning this analyte can be calculated for well trained male athletes.

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