Repeated Bout Effect and Indirect Biomarkers After Eccentric Exercise Induced Muscle Damage

11 February 2026

Information for Participants and Consent to Participate in Research

The purpose of this study is to investigate changes in markers of exercise-induced muscle damage following an acute and a subsequent eccentric exercise of the knee joint extensors performed on an isokinetic dynamometer. During the study, the following measurements/assessments will be carried out:

Eccentric exercise (knee extensors)

Eccentric exercise of the knee joint extensors will be performed on an isokinetic dynamometer. The exercise will consist of 5 sets of 10 repetitions (knee extensions), while the angular velocity will be set at 60°/s.

Assessment of peak torque

An assessment of isokinetic peak torque will be performed on an isokinetic dynamometer. The assessment will consist of one set of 8 repetitions for familiarization and one set of 5 maximal-effort repetitions.

Possible discomforts

Muscle fatigue and cardiovascular strain. In rare cases, there is a possibility of fainting; in such a case, the appropriate equipment (examination bed) is available, while the tests will be conducted at times when a doctor or nurse will be present at the faculty facilities. To reduce discomfort, a warm-up will be performed (low-intensity exercise on a cycle ergometer for 7 minutes and stretching exercises of the main muscle groups) and a cool-down (stretching). Approximately 2 days after the eccentric exercise, the sensation of muscle pain during movement (delayed onset muscle soreness) in the exercised muscle groups will peak. The sensation of pain will gradually subside over the following days.

Blood sampling

On the days of the assessments, a blood sample of 10 ml will be taken from the vein on the inner side of the elbow joint (basilic vein). A total of four (4) blood draws will be performed during the study.

Possible discomforts

Mild local pain and the possibility of bruising (hematoma) in the area. In rare cases, there is a possibility of fainting; in such a case, the appropriate equipment (examination bed) is available. Blood sampling will be performed by a doctor at the facilities of the School of Physical Education and Sport Science.

Muscle oxygenation

On the days of the assessments, a NIRS device will be placed on the knee extensors for monitoring muscle oxygenation and will be there for the whole period of the data collection.

Possible discomforts

None.

It is important that you do not withhold any information you know that relates both to your current health status and to any issue that may arise during the measurements. The results of the measurements are confidential and will be used only by the research team, while any publication of results will be anonymous. A necessary condition for your participation in the study is examination and written approval by an internal medicine physician stating that you can participate in maximal-intensity exercise without risk to your health.

Consent Statement

I declare that I have understood the procedure of the above assessments and the risks involved and I wish to participate in the research program. I also understand my right to withdraw from

the above tests at any time I wish, without any obligation to provide any explanation to the researchers.

Date ……/……/ 2025

Participant’s name:
Signature:

Researcher’s name:
Signature:

Witness’s name:
Signature:

Study design
Experimental groups performed 2 eccentric exercise sessions unilateral sessions using the knee extensors on an isokinetic dynamometer (Biodex, 4 pro, Shirley, NY, USA). The second eccentric exercise session was performed 3, 6, 9 or 12 weeks after the initial exercise bout. A number of physiological and biochemical markers related to muscle damage were assessed. Specifically, physiological markers of muscle function and damage [pain-free range of motion (ROM), delayed onset muscle soreness (DOMS), and isokinetic peak torque] and biochemical markers of muscle damage (creatine kinase; CK) and inflammation (C-reactive protein; CRP) were measured prior to as well as 48 and 72 h post each exercise session.

Statistical analysis
The distribution of all dependent variables will be examined by the Kolmogorov–Smirnov test. The anthropometric characteristics of the 4 groups at baseline will be examined using an unpaired Student's t test. A two-way repeated-measures ANOVA test [group (3, 6, 9 and 12 weeks) × time (pre-exercise
and 48- and 72-hours post-exercise)] will be performed to compare muscle function and damage biomarkers between the groups and to compare the effect of each exercise mode on muscle damage biomarkers in each week of intervention. When a significant interaction will be obtained, pairwise comparisons will be performed through the Sidak test that counteract the problem of multiple comparisons. When sphericity will be violated, the Greenhouse–Geisser correction will be applied. Data will be presented as mean ± standard deviation (SD) and the level of significance will be set at a = 0.05.
The SPSS version 26.0 will be used for all analyses (SPSS Inc., Chicago, IL).