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Systemic cytokine response to three bouts of eccentric exercise

Stephen M. Cornish [a,*], Steven T. Johnson [b]

[a] Faculty of Kinesiology & Recreation Management, University of Manitoba, Winnipeg, Manitoba, Canada
[b] Centre for Nursing and Health Studies, Athabasca University, Athabasca, Alberta, Canada

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ABSTRACT

This research examined the changes in inflammatory cytokines interleukin 6 (IL-6), IL-1β, IL-10, as well as muscle force, muscle soreness, thigh circumference, and range of motion in response to 3 bouts of eccentric knee extension. Ten males were recruited to participate. The participants performed eccentric exercise on 3 consecutive days on the knee extensors on the right leg separated by 24 h. Participants performed 6 sets of 10 repetitions of isokinetic eccentric knee extension at 120° per second. Blood was sampled before and after each exercise bout and 24 h after the final exercise bout. Muscle isometric force, delayed onset muscle soreness (DOMS), thigh circumference, and range of motion were evaluated before and after each exercise bout and 24 h after the final exercise bout. There were no statistically significant differences noted for the changes in isometric strength, thigh circumference, and range of motion, or IL-6 over the 4 days (all $p > 0.05$). On the second day and third day there was a significant increase noted in DOMS as compared with baseline ($p < 0.05$). These results suggest that 3 consecutive days of eccentric exercise results in DOMS but does not produce a sustained systemic inflammatory reaction or changes in muscle function.

1. Introduction

High intensity eccentric exercise is known to cause skeletal muscle damage and micro-structural changes to muscle fibers with an associated inflammatory response [1,2]. This skeletal muscle damage has been shown to limit muscle strength and performance [3]; however, there is a phenomenon known as the repeated bout effect whereby a previous eccentric exercise bout seems to promote an adaptation that will limit muscle damage, inflammation, and loss of function if a similar bout of exercise is performed after a recovery period [4]. There are a number of theories surrounding the mechanism whereby the repeated bout effect functions including the mechanical theory, the neural theory, and the cellular theory [4]. Within the cellular theory there is a mechanism which proposes that there is less of an inflammatory reaction following the second bout of exercise and this may be the reason for the maintenance or return of muscle strength and function to a greater degree following the second bout as compared to the first bout of eccentric exercise [4].

Even though the mechanism whereby the repeated bout phenomenon occurs is not known, previous research has evaluated the cytokine response to eccentric exercise utilizing the repeated bout effect model. Prior research does indicate an increase in mRNA expression of interleukin-6 and interleukin-8 following a downhill treadmill run [5]. Also, neutrophils, macrophages, and IL-1β do accumulate in skeletal muscle after an acute bout of eccentric exercise [1,6,7]. Further data suggests a systemic inflammatory response to high intensity eccentric resistance exercise where there is an increase in IL-6 and IL-10 [8]. After completing 2 bouts of downhill running, with 2 weeks of rest in between bouts, Smith et al. [9] demonstrated a decrease in IL-6 and an increase in IL-10 after the second bout of exercise when compared to the first in untrained males. Conversely, knee extensor eccentric exercise separated by 3 weeks did not reveal any change in the mRNA or blood IL-6 response to the exercise between the first and second bout [10]. Further, when compared to pre-exercise concentrations, two bouts of eccentric elbow flexion, separated by 4 weeks, demonstrated a decrease in TNF-α and IL-8 after the first bout of exercise and an increase in IL-10 and decrease in IL-8 following the second bout of exercise [11]. Thus, there does not seem to be any clear consensus as to the cytokine response to eccentric exercise during a repeated bout of exercise. The variation in the response of cytokines noted above is likely related to the type or amount of muscle activated during the eccentric exercise and may be related to the time interval between the exercise bouts.

Eccentric exercise and the associated muscle damage will result in delayed onset muscle soreness (DOMS), reduced range of motion, decreased muscular strength, and increased fluid accumulation (edema) in the affected muscle [1]. All of these effects will likely result in reduced performance in a subsequent exercise bout. Nevertheless, the majority of research looking at the

* Correspondence to: Faculty of Kinesiology & Recreation Management, University of Manitoba, Winnipeg, Manitoba, Canada.
E-mail address: Stephen.Cornish@umanitoba.ca (S.M. Cornish).

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repeated bout effect has been done in studies with 1–3 weeks of recovery between the consecutive bouts [1,9–13]. This is not typical of an athlete's training regime as many times athletes repeat training techniques (such as plyometrics) within 2–3 days. Also, tournament play may require athletes to perform eccentric muscle actions associated with the sport on the same or subsequent days. Thus, a further examination of the inflammatory response associated with repeated bouts of eccentric exercise performed on consecutive days will provide important new information regarding the effects of eccentric exercise in athletes. The amount of time given for rest and recovery following the initial bout of exercise may play a key role in the inflammatory response to exercise. Chen and Hsieh [14] reported an acute increase (after 1 day) in IL-1β and IL-6 after 30 maximal repetitions of the elbow flexors at $60^\circ \times s^{-1}$ with 24 h between bouts. This research was done in a relatively small group of muscles and therefore, it would be interesting to evaluate the effects that minimal rest between exercise bouts has on the systemic cytokine response to repeated bouts of eccentric exercise in a larger muscle group. Few studies have been conducted that evaluated short time intervals of rest between repeated bouts of eccentric exercise on consecutive days therefore, the main purpose of this study was to evaluate the systemic cytokine response to 3 repeated bouts of knee extensor eccentric exercise separated by a 24 h rest period in healthy young men. The main hypothesis was that there would be less of a pro-inflammatory reaction and more of an anti-inflammatory reaction to the eccentric exercise protocol on days 2 and 3 as compared to day 1.

2. Material and methods

2.1. Participants

This study recruited healthy young males between the ages of 18 and 30 years. Participants were recruited by placing posters around the university campus and word of mouth. Participants were excluded from the study based on the following criteria: (1) consumption of anti-inflammatory medication (non-steroidal anti-inflammatory drugs, aspirin, or other immunosuppressive medication) within the past month, (2) diagnosed with an inflammatory disease (rheumatoid arthritis, inflammatory bowel disease etc.), (3) had sustained a recent injury, (4) had an illness (infection) within the past 2 weeks or, (5) were taking vitamin, mineral, or other nutritional supplements.

The sample size estimated for this study was based on previous research [9] where IL-6 concentration was decreased by 50% immediately following the second bout of eccentric exercise when compared to the first bout of eccentric exercise. With a power of 80%, an alpha level of 0.05, and an expected attrition rate of 25%, 10 participants were recruited which accounted for the expected attrition rate (Statistica version 7, StatSoft Inc., Tulsa, Oklahoma, USA).

2.2. Procedures

A single group design was used to evaluate the effects of three separate eccentric exercise bouts on inflammatory markers in males. Dependent variables included: serum concentration of IL-1β, IL-6, and IL-10 as well maximal knee extensor isometric torque, delayed onset muscle soreness, range of motion of the knee joint, and upper leg circumference. Participants were familiarized with all the measurement protocols and the exercise intervention protocol and performed the initial test of dependent variables on the first visit to the laboratory. In total, the participants visited the laboratory 4 times; 3 times for the exercise intervention/testing as well as one time 24 hh after the final bout of eccentric exercise to measure the dependent variables again (see Fig. 1). This study was approved by the Human Participant Research Committee at the University of Lethbridge.

2.3. Maximal muscle force

Maximum isometric knee extension force was measured on the right leg of each individual using an isokinetic dynamometer (CSMi Humac NORM, Stoughton, Maryland, USA). Participants assumed a seated position and were secured to the chair by stabilizing belts placed across the chest, over the lap, and on the distal one-third of the thigh on the tested leg. Participants sat against a back support with an 80° angle of the hip flexors. The rotational axis of the dynamometer was placed coaxial to the knee axis (lateral femoral epicondyle) and the lever arm of the dynamometer was secured to the distal shin by a strap. The knee angle was fixed at 60° of knee flexion for the isometric contraction with 0° being full knee extension. Participants were asked to exert maximal isometric force against the dynamometer for 10 s and they were verbally encouraged to do so during the entire test. The participants were given a 60 s rest and then the procedure was repeated two more times with the highest torque in Newton meters (N hm) recorded. Maximal isometric knee extension force was measured immediately before and after the initial bout of eccentric exercise, before and after exercise bout 2 and 3, and 24 h after the final exercise bout.

2.4. Eccentric exercise intervention

The intervention involved three sessions of maximal eccentric exercise of the knee extensors of the right leg separated by a 24 h recovery period. During the 24 h recovery period participants were asked to refrain from any form of physical activity or exercise. Participants performed 6 sets of 10 repetitions of maximal isokinetic eccentric knee extension (CSMi Humac NORM, Stoughton, Maryland, USA) at $120^\circ \times s^{-1}$ with 1 min of rest between each set on each day. This protocol has been used previously to induce muscle damage [13]. Participants were seated and secured to the dynamometer chair as described above but the range of motion of the dynamometer was set between 0° (full knee extension) and 90° (knee flexion) for the eccentric knee extension exercise. The participants were instructed to resist with maximal force against the dynamometer arm as it moved their knee from extension to flexion. The dynamometer motor was used to passively bring the leg back up to the starting position after each repetition was completed. All participants were verbally encouraged to exert maximal resistance to the dynamometer throughout each session.

2.5. Delayed onset muscle soreness

A visual analog scale (VAS) was used to assess delayed onset muscle soreness (DOMS). The instrument consists of a 100 mm long line with the words "no pain" on the left hand side and "unbearable pain" on the right hand side of the line. Participants were asked to draw a vertical line on the scale to rate their current level of pain in their right leg. A pain-rating index was calculated by measuring the distance of the mark from the left hand side of the diagram in millimeters [15]. Pain was assessed before and after each exercise bout as well as 24 h after the final bout with the participants in a standing position.

2.6. Thigh circumference

Thigh circumference was measured as an indicator of any edema that may have occurred due to the eccentric exercise intervention. To measure thigh circumference, the halfway distance between the greater trochanter of the femur and the lateral