EFFECTS OF CONCENTRIC AND ECCENTRIC CONTRACTIONS ON EXERCISE-INDUCED MUSCLE INJURY, INFLAMMATION, AND SERUM IL-6

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ABSTRACT

EFFECTS OF CONCENTRIC AND ECCENTRIC CONTRACTIONS ON EXERCISE-INDUCED MUSCLE INJURY, INFLAMMATION, AND SERUM IL-6. Darryn S. Willoughby, Clesi VanEnk, Lemuel Taylor. JEPonline. 2003;6(4):8-15. The present study determined the effects of concentric and eccentric contractions on muscle function, injury, swelling/inflammation, and serum skeletal muscle troponin-I (sTnI), creatine kinase (CK), cortisol (CORT), and interleukin-6 (IL-6). Eight untrained males performed one exercise bout with each leg, separated by three weeks. One bout consisted of 7 sets of 10 repetitions of eccentric contractions of the knee extensors at 150% of the concentric 1-RM (ECC) while the other bout consisted of 7 sets of 10 repetitions of concentric contractions at 75% 1-RM (CON). The legs used and the bouts performed were randomized to prevent order effects. Five days prior to each exercise bout, baseline measurements were taken for muscle strength. Immediately prior to each bout, a venous blood sample and measurements of thigh circumference (CIRC), total body water (TBW), knee range of motion (ROM), and perceived muscle soreness (SORE) were obtained. At 6, 24, and 48 hr post-exercise, all aforementioned measurements and blood samples were obtained for both bouts. Data were analyzed with 2 X 4 repeated measures ANOVA (p<0.05). ECC produced greater muscle injury, evidenced by peak decreases of 30% in strength and increases of 307%, 228%, 66%, 78%, and 170%, respectively, for SORE, CK, sTnI, CORT, and IL-6 (p<0.05). However, the 5%, 6%, and 5% increases in CIRC, TBW, and ROM were not significant (p<0.05). For CON, respective peak decreases in strength and ROM of 12% and 2%, and increases of 3% and 3% for CIRC and TBW were not significant. However, the respective increases of 173%, 143%, 31%, 30%, and 56% for SORE, CK, sTnI, CORT, and IL-6 were significant (p<0.05). Our results suggest that eccentric contractions induce greater muscle injury than concentric contractions; however, the magnitude of muscle injury seems to be the primary mitigating factor for increasing serum IL-6 rather than muscle inflammation.

Key Words: Acute-phase immune response, Cytokine, Inflammation
INTRODUCTION

Compared to concentric contractions, dynamic muscle contractions involving a significant eccentric (forced-lengthening) component are thought to produce a greater degree of muscle injury resulting in exacerbated reductions in muscle strength and range of motion (ROM), while concomitantly producing muscle soreness and swelling/inflammation. These consequences are also often associated with increased levels of serum markers of muscle injury [e.g., creatine kinase (CK), skeletal muscle troponin-I (sTnI)], inflammation [e.g., interleukin-6 (IL-6)], and endocrine markers of physiological stress [cortisol (CORT)].

Eccentric contractions apparently produce more muscle damage than concentric contractions. The cause of this discrepancy is multifaceted. For example, eccentric contractions have been shown to invoke 40% less EMG activity compared to concentric contractions (1), which can be interpreted as reduced motor unit recruitment during the eccentric phase of contraction. This indicates that during an eccentric contraction a smaller cross-sectional area takes on an equivalent load as that which was handled in the concentric contraction (2). In addition, eccentric contractions result in a large degree of myofibrillar damage due to overstretching of the sarcomeres within the myofibrils, resulting in the disruption to the sarcolemma and degradation of the contractile machinery; these events eventually lead to decrements in muscle strength due to excitation-contraction coupling dysfunction (3).

The reductions in muscle strength and ROM as a result of eccentric contractions are often accompanied by soreness (SORE), swelling, and stiffness (4,5). The level of soreness and swelling/inflammation is associated with the degree of injury induced and increases in limb circumference after eccentric exercise are related to inflammatory swelling (6). Furthermore, exercise-induced muscle injury is thought to instigate an “acute-phase” local inflammatory response. This response is associated with the release of pro-inflammatory cytokines [e.g., interleukin-6 (IL-6) and tumor necrosis factor] at the site of muscle injury in order to facilitate healing of the tissue (6) and may also play a role in increasing the levels of serum CORT (7). The cytokine, IL-6, is thought to be inflammation responsive and play an integral role in controlling the acute-phase immune response that accompanies exercise-induced muscle injury. We have previously shown eccentric contractions to induce muscle injury (8,9) and increase serum CORT (10), and that repeated bouts of eccentric exercise attenuate the magnitude of muscle injury without any effect on serum IL-6, suggesting IL-6 to possibly be responsive to muscle injury and stress rather than inflammation (8). Also, IL-6 has been shown to increase in response to dynamic muscle contractions (11); however, the elevations in IL-6 in response to eccentric exercise are normally observed to be greater than concentric exercise suggesting that the acute-phase response imparted by IL-6 seems to be associated with the muscle injury resulting from this type of muscle activity (12). In addition, it has been suggested that the elevated plasma IL-6 levels following eccentric exercise may induce proteolysis and prolong muscle injury (13,14).

Therefore, the purpose of the present study was to determine the effects of concentric and eccentric contractions of the knee extensors on measures of muscle function (strength, ROM, and SORE), stress (CORT), injury (serum CK and sTnI), swelling [thigh circumference (CIRC) and total body water (TBW)], and inflammation (serum IL-6). We hypothesized that the greater severity of muscle injury resulting from eccentric contractions would be evident from greater increases in serum sTnI and CK and decrements in strength, thereby resulting in higher levels of muscle soreness, inflammation, and serum CORT and IL-6 levels compared to concentric contractions.

METHODS

Subjects

Eight untrained, recreationally active males were recruited to participate in the study. The subjects were untrained from the standpoint that they had not engaged in consistent weight training for 3 months prior to the study; however, all were recreationally active. The eight subjects had a mean age of 20.6 ± 1.3 years, height of
72.6 ± 2.1 inches, and body mass of 170.9 ± 18.5 kg. Before participating, each subject completed a medical history questionnaire, was informed of the experimental protocol, and signed a university-approved informed consent form. Subjects with contraindications to exercise as indicated by the American College of Sports Medicine were not allowed to participate.

**Muscle Strength**

Based on our previous guidelines (8-10), each subject underwent strength testing to determine the concentric strength of the knee extensors using a trial-and-error method of assessing the one repetition maximum (1-RM) on a leg extension machine (Universal, Cedar Rapids, IA). Strength tests were performed 5 days prior to and at 6, 24, and 48 hr after each exercise bout. Initial strength tests as well as the damage protocol were performed on the same apparatus. In order to prevent fatigue as a result of excessive trials (i.e., >5 trials) during 1-RM testing, based on our previous work, a goal of only five trials was set for all 1-RM testing sessions throughout the study (8-10). All subjects were able to obtain their 1-RM within 5 trials and the average (±SD) number of trials for all subjects over the eight 1-RM testing sessions was 4.2 ±0.7.

**Range of Motion (ROM)**

As an indicator of passive muscle stiffness caused by either eccentric or concentric contractions, knee ROM was determined using a goniometer (Jamar EZ-Read, Clifton, NJ) immediately prior to each exercise bout and at 6, 24, and 48 hr after each bout. Subjects assumed a supine position with the femur 90 degrees to the hip joint and were asked to extend at the knee joint. The head of the femur, lateral epicondyle of femur, and lateral malleolus were used as bony landmarks and range of motion was assessed as the femur being at 0 degrees (15).

**Thigh Circumference (CIRC) and Total Body Water (TBW)**

As an indicator of muscle inflammation caused by either eccentric and concentric contractions, muscle swelling/inflammation was determined by measurements of CIRC and TBW. Measurements of CIRC were performed utilizing a Gulick anthropometric tape (Model J00305, Lafayette Instruments, Lafayette, IN) at mid thigh immediately prior to each exercise bout and at 6, 24, and 48 hr after each bout. Mid thigh was considered ½ the femur length (as measured from head of femur to lateral epicondyle). Once ½ femur length was determined a semi-permanent mark was drawn an equivalent distance proximal the superior border of the patella (16). Measurements of TBW were performed by way of bioelectrical impedance with a Tanita BF-350e body composition analyzer (Tanita UK Ltd., Yiewsley Middlesex, UK).

**Perception of Soreness (SORE)**

Soreness was assessed along a 10-inch scale (0 = no soreness, 10 = extreme soreness). Subjects rated their level of SORE immediately prior to each exercise bout and at 6, 24, and 48 hr after each bout by drawing an intersecting line across the continuum line extending from 0-10. The distance of each mark was measured from zero and the measurement utilized as the perceived SORE level (8-10).

**Blood Sampling**

Venous blood samples consisted of approximately 10 mL of blood drawn from the antecubital vein using a vacutainer apparatus immediately prior to each bout and at 6, 24, and 48 hr following each bout. Blood was centrifuged for 10 minutes and serum was extracted and then stored at a temperature of -20 °C.

**Exercise bouts**

Each subject underwent two separate muscle injury-inducing exercise sessions. One session involved concentric contractions only of the knee extensors (CON) and the other session involved eccentric contractions only of the knee extensors (ECC). Each exercise bout was separated by three weeks and exercise sessions alternated the leg and type of exercise to avoid the repeated bout effect. For example, if the first exercise session incorporated the left leg and eccentric contractions then the subsequent session 3 weeks later utilized the right leg and concentric contractions. The type of muscle contractions as well as which leg was used were both randomized to control for order effects. Both CON and ECC exercise sessions followed identical protocols. Each exercise bout employed 7 sets of 10 repetitions. However, the ECC bout involved eccentric contractions of the knee extensors using 150% of the concentric 1-RM (8-10,17). In an attempt to standardize for the amount of repetitions across bouts, based on the repetition continuum, a relative intensity of 75% was chosen for the CON bout. This is based on the premise that 75% 1-RM corresponds to a 10-RM (18). For the ECC bout, study investigators raised the weight prior to each repetition, whereas in the CON bout, study investigators lowered
the weight after each repetition. Both bouts began with two warm-up sets of 10 repetitions at 50% of each subject’s 1-RM. For both exercise bouts, each repetition was approximately 2-3 seconds in duration, each repetition was separated by 15-sec rest interval, and each set was separated by a 3-min rest interval.

**Serum Protein Quantitation**

The serum levels of sTnI (a marker of muscle injury), cortisol (a marker of physiological stress), and IL-6 (a marker of muscle inflammation) were determined with an enzyme-linked immunoabsorbent assay (ELISA) (8-10) using primary monoclonal antibodies for anti-sTnI (Avanced ImmunoChemical, Long Beach, CA), anti-CORT, (Affiniti Research Products, Mamhead, UK), and anti-IL6 (Research Diagnostics Inc., Flanders, NJ). The secondary antibody (ICN Biomedical, Aurora, OH) involved immunoglobulin-G (IgG) conjugated to the enzyme horseradish peroxidase. The assays were run in duplicate and the protein concentrations were determined at an optical density of 450 nm with a microplate reader (Bio Rad, Hercules, CA). Intra-assay coefficients of variation were 4.74%, 4.87%, and 3.97%, respectively, for sTnI, CORT, and IL-6.

The levels of serum CK were determined using a clinical diagnostic kit (Pointe Scientific, Lincoln Park, MI) based on the manufacturer’s guidelines. Briefly, the principle of the kit is based on a series of enzymatic reactions whereby CK catalyzes the reversible phosphorylation of ADP, in the presence of creatine phosphate, to form ATP and creatine. The series of subsequent reactions form NADH, and the rate of NADH formation, measured at 340 nm, is directly proportional to serum CK activity.

**Statistical Analyses**

Statistical analyses on the raw data of each criterion variable were performed by utilizing separate 2 x 4 [Bout (eccentric, concentric) x Test (pre-exercise and 6, 24, 48 hr post-exercise)] factorial analyses of variance (ANOVA) with repeated measures for each independent variable. In the event of a significant Bout x Test interaction, between-group differences were determined using the Student Neuman-Keuls Post Hoc Test. All statistical procedures were performed using SPSS 11.0 software and a probability level of p<0.05 was adopted throughout. Data are presented as mean±SD.

**RESULTS**

Tables 1 presents the data of each of the criterion variables for the CON and ECC exercise bouts at pre-exercise and at 6, 24, and 48 hr post-exercise. In addition, the percent change from pre-exercise for each criterion variable is also presented.

**Maximum Dynamic Strength**

In regard to muscle strength, a significant Bout x Test interaction (F(2,7)=5.82, p=0.032) was detected. Results showed that ECC caused greater decrements in strength than CON. Post-hoc analyses indicated that for ECC the decrease in strength was significantly greater (p<0.05) at 24 hr post-exercise.

**Muscle Soreness**

In regard to muscle soreness, there was a significant Bout x Test interaction (F(2,7)=7.43, p=0.01) detected. Results showed that ECC caused greater amounts of muscle soreness than CON. Post-hoc analyses indicated that the increase in soreness was significantly greater (p<0.05) at 24 and 48 hr post-exercise for ECC and 24 hr post-exercise for CON.

**Range of Motion, Thigh Circumference, and Total Body Water**

There were no significant differences observed for ROM, CIRC, and TBW (p>0.05).

**Serum Creatine Kinase and sTnI Content**

A significant Bout x Test interaction was observed for serum CK (F(3,7)=4.44, p=0.01). Results showed that ECC caused greater increases in CK than CON. Post-hoc analyses indicated that ECC experienced significantly greater (p<0.05) increases in CK at 6, 24, and 48 hr while CON only experienced significant increases in CK at 24 hr post-exercise.

A significant Group x Test interaction was observed for serum sTnI (F(3,5)=3.86, p=0.01). Results showed that ECC caused greater increases in sTnI than CON. Post-hoc analyses indicated that ECC experienced...
significantly greater (p<0.05) increases in sTnI at 24 and 48 hr while CON only experienced significant increases in sTnI at 24 hr post-exercise.

Serum Cortisol and IL-6 Content
No significant interaction was observed for serum CORT (p>0.05); however, a significant Group x Test interaction was observed for serum IL-6 (F(3,5)=5.31, p=0.001). Results showed that ECC caused greater increases in IL-6 than CON. Post-hoc analyses indicated that ECC experienced significantly greater (p<0.05) increases in IL-6 at 6, 24, and 48 hr while CON only experienced significant increases in IL-6 at 24 hr post-exercise.

Table 1. Mean?SD data for the dependent variables in response to CON and ECC contractions for the three times of recovery.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Pre-Exercise</th>
<th>6 hr</th>
<th>24 hr</th>
<th>48 hr</th>
<th>Peak % Change</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Strength (kg)</strong> (Bout x Test, p&lt;0.05)</td>
<td></td>
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<tr>
<td>CON</td>
<td>44.34±6.89</td>
<td>41.67±7.93</td>
<td>39.13±4.98</td>
<td>42.76±5.23</td>
<td>-11.75</td>
</tr>
<tr>
<td>ECC</td>
<td>43.20±7.01</td>
<td>40.54±6.65</td>
<td>30.06±5.47 †</td>
<td>40.67±4.25</td>
<td>-30.28</td>
</tr>
<tr>
<td><strong>SORE</strong> (Bout x Test, p&lt;0.05)</td>
<td></td>
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<td></td>
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</tr>
<tr>
<td>CON</td>
<td>2.12±1.78</td>
<td>2.49±1.06</td>
<td>5.79±0.95 †</td>
<td>2.34±0.77</td>
<td>173.11</td>
</tr>
<tr>
<td>ECC</td>
<td>2.05±1.19</td>
<td>3.65±2.18</td>
<td>8.34±1.19 †</td>
<td>3.73±0.95 ‡</td>
<td>306.82</td>
</tr>
<tr>
<td><strong>CIRC (cm)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>CON</td>
<td>54.12±1.96</td>
<td>55.44±2.73</td>
<td>55.86±2.61</td>
<td>55.12±2.18</td>
<td>3.21</td>
</tr>
<tr>
<td>ECC</td>
<td>54.64±1.78</td>
<td>55.58±1.65</td>
<td>57.46±1.87</td>
<td>55.35±1.94</td>
<td>5.16</td>
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<tr>
<td><strong>TBW (kg)</strong></td>
<td></td>
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<td></td>
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<tr>
<td>CON</td>
<td>48.37±12.55</td>
<td>48.85±12.71</td>
<td>50.05±11.64</td>
<td>47.32±11.79</td>
<td>3.49</td>
</tr>
<tr>
<td>ECC</td>
<td>45.40±6.20</td>
<td>45.12±7.87</td>
<td>48.21±7.42</td>
<td>46.60±7.05</td>
<td>5.95</td>
</tr>
<tr>
<td><strong>ROM (degrees)</strong></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>CON</td>
<td>147.21±12.85</td>
<td>145.98±12.87</td>
<td>144.32±13.91</td>
<td>148.54±12.70</td>
<td>-1.96</td>
</tr>
<tr>
<td>ECC</td>
<td>148.18±7.79</td>
<td>146.35±7.32</td>
<td>140.06±8.21</td>
<td>146.21±9.83</td>
<td>-5.47</td>
</tr>
<tr>
<td><strong>sTnI (ng/ml)</strong> (Bout x Test, p&lt;0.05)</td>
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<tr>
<td>CON</td>
<td>69.26±25.05</td>
<td>78.38±38.26</td>
<td>91.05±42.12 †</td>
<td>82.47±42.56</td>
<td>31.46</td>
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<tr>
<td>ECC</td>
<td>71.34±26.58</td>
<td>89.65±43.22</td>
<td>128.55±40.49 †</td>
<td>98.30±34.32 ‡</td>
<td>80.19</td>
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<tr>
<td><strong>CK (U/ml)</strong> (Bout x Test, p&lt;0.05)</td>
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</tr>
<tr>
<td>CON</td>
<td>93.07±30.05</td>
<td>152.68±46.42</td>
<td>226.55±25.92 †</td>
<td>138.13±21.22</td>
<td>143.41</td>
</tr>
<tr>
<td>ECC</td>
<td>104.60±26.81</td>
<td>248.72±65.57 *</td>
<td>342.84±65.56 †</td>
<td>198.05±61.24 ‡</td>
<td>227.76</td>
</tr>
<tr>
<td><strong>CORT (ug/dl)</strong></td>
<td></td>
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<td></td>
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<td></td>
</tr>
<tr>
<td>CON</td>
<td>13.25±1.86</td>
<td>15.07±2.48</td>
<td>17.24±2.96</td>
<td>15.34±2.89</td>
<td>30.11</td>
</tr>
<tr>
<td>ECC</td>
<td>13.16±1.97</td>
<td>17.66±1.37</td>
<td>21.51±3.26</td>
<td>16.44±2.83</td>
<td>63.44</td>
</tr>
<tr>
<td><strong>IL-6 (pg/ml)</strong> (Bout x Test, p&lt;0.05)</td>
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<td></td>
</tr>
<tr>
<td>CON</td>
<td>46.40±25.40</td>
<td>49.10±21.50</td>
<td>72.40±29.60 †</td>
<td>46.50±21.10</td>
<td>56.25</td>
</tr>
<tr>
<td>ECC</td>
<td>37.50±23.50</td>
<td>66.30±26.90 *</td>
<td>101.11±23.10 †</td>
<td>59.50±31.30 ‡</td>
<td>169.25</td>
</tr>
</tbody>
</table>

* Significantly different at 6 hr post-exercise, † significantly different at 24 hr post-exercise, ‡ significantly different at 48 hr post-exercise (p<0.05).

DISCUSSION

Results from the present study support our hypothesis that the greater severity of muscle damage induced by eccentric muscle contractions are more effective at increasing muscle soreness an serum IL-6 than concentric contractions. However, the present results did not support our hypothesis that eccentric contractions would
result in greater increases in serum CORT, degree of muscle inflammation (increased CIRC and TBW) and muscle stiffness (decreased knee ROM).

In line with our previous work (8-10), here we observed significant decrements in dynamic muscle strength of 30% for ECC whereas CON only underwent modest decrements in strength of 12%. Due to the combination of motor unit de-recruitment and decreased EMG activity during eccentric contractions, the early strength loss due to muscle injury from eccentric exercise apparently results from a failure of excitation-contraction uncoupling processes, likely due to disruption of the contractile machinery and a subsequent loss of myofibrillar protein (19). In light of this possible disruption to the sarcolemma, with ECC we observed significant serum increases in the myofibrillar proteins CK and sTnI (both markers of muscle injury) of 228% and 66%, respectively, compared to the corresponding increase of 143% and 31% for CON. Therefore, it is apparent that ECC resulted in a greater magnitude of muscle injury than CON. Consequently, fast-twitch muscle fibers have been shown to be more susceptible to eccentric contraction-induced muscle injury (20) possibly due to sub-optimal matching of protein properties and physiological demands, leading to sarcomere instability (21). Incidentally, the human quadriceps femoris contains a high percentage of Type II muscle fibers. For example, the vastus lateralis contains approximately 57% Type II muscle fibers (22).

It is thought that increases in muscle swelling/inflammation along with myofiber disruption following eccentric exercise results in increases in passive muscle stiffness, thereby decreasing ROM at the affected joint (23). However, it has been shown that muscle swelling does not account for the increase in passive stiffness within the first 48 hr following eccentric exercise (24). The present results reveal that ECC resulted in a larger increase in CIRC (5%) and TBW (6%) and decreased ROM (5%) when compared to the increased CIRC (3%) and TBW (3%) and decreased ROM (2%) for CON; however, none of these changes were statistically significant for ECC or CON, and in both cases the decreased ROM did not appear to be predicated on the increased CIRC.

Previous results have shown that eccentric exercise had no significant effect on the levels of serum CORT (25, 26). However, we have recently shown serum CORT to be elevated after an initial bout of eccentric exercise but was attenuated after a repeated eccentric exercise bout three weeks later (10). Also, there are also data indicating that IL-6 induces increases in serum CORT (7). Results from the present study suggest that eccentric exercise does not have a pronounced effect on serum CORT compared to concentric exercise, and that serum CORT is not contingent on serum IL-6 levels. Although, our data should be interpreted cautiously since CORT is a stress hormone that is released in a pulsatile manner. The body's level of serum CORT is subject to diurnal variations in which normal concentrations vary throughout a 24-hour period. Cortisol levels in normal individuals are highest in the early morning at around 6-8 am and are lowest around midnight (27). Therefore, as a result of our blood sampling periods it is conceivable that we may have missed the window of time in which peak CORT levels due to either concentric or eccentric exercise occurred.

Previous results have indicated a relationship between eccentric exercise-induced muscle injury to contractile proteins and inflammation, and that muscle soreness is associated with inflammation but not with muscle injury (28). However, our results demonstrate the contrary for eccentric exercise showing no significant effects on CIRC and TBW (indicators of muscle swelling/inflammation), whereas SORE was significantly increased 307% (p<0.05). Consequently, CON resulted in an elevated level of SORE of 173% (p<0.05) but was less than ECC. Therefore, our results suggest that ECC results in greater perceptions of SORE than CON and that SORE seems to be contingent on the severity of muscle injury rather than the level of muscle swelling/inflammation. In light of our results, however, SORE has been suggested to be a poor reflector of eccentric exercise-induced muscle injury and inflammation, and changes in markers of muscle damage and inflammation are not necessarily accompanied by SORE (29).
It has been suggested that an eccentric exercise-induced increase in IL-6 is associated with muscle damage. It is also known that with eccentric exercise the IL-6 kinetics differ from those of concentric exercise (12). Moreover, with concentric exercise, IL-6 levels increase during exercise and decrease upon the cessation of exercise (11). However, a modest, prolonged increase in IL-6 is considered to occur after eccentric exercise (12), suggesting that IL-6 may be indicative of the instigation of repair mechanisms after muscle injury. Our present results support the contention that serum IL-6 levels are dependent on contraction type. We observed only a modest increase in IL-6 of 56% with CON; however, ECC produced significant increases in IL-6 of 170% (p< 0.05). Additionally, CIRC did not significant increase with either ECC or CON.

Based on the results presented in this study, we conclude that the severity of muscle injury produced by eccentric muscle contractions resulted in a greater decrement in muscle strength than concentric muscle contractions. Furthermore, the severity of muscle injury induced by eccentric contractions result in greater increases in soreness and serum IL-6, independent of muscle inflammation and stiffness and serum CORT, suggesting that the magnitude of muscle injury, rather than inflammation, is a more likely stimulus in initiating the pro-inflammatory cytokine response of IL-6.

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