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1 **ENDURANCE MARKERS ARE RELATED WITH LOCAL NEUROMUSCULAR**
2 **RESPONSE FOR THE INTACT BUT NOT FOR THE ACL RECONSTRUCTED LEG**
3 **DURING HIGH INTENSITY RUNNING**

4

5 Short title: Endurance correlates of EMG response in ACLR athletes

6

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1 **ABSTRACT**

2 **AIM:** It has been demonstrated that the local neuromuscular response during high intensity
3 exercise has a strong relationship with endurance markers. However a diminished
4 neuromuscular response has been reported for the operated leg in athletes having undergone
5 anterior cruciate ligament reconstruction (ACLR). The purpose of the present study was to
6 examine the relationships between endurance markers and the EMG response during high
7 intensity running in ACLR athletes. **METHODS:** Fourteen ACLR soccer players underwent
8 a GXT test to volitional exhaustion and a 10-min bout of high intensity running. During the
9 10-min bout, EMG data were recorded at the 3rd and 10th minute from the vastus lateralis
10 bilaterally using a telemetric system. The final EMG levels were expressed as a percentage of
11 the initial values. Pearson moment product correlations were used to assess the relationship
12 between the endurance markers of VO₂max, velocity at lactate threshold (vLT), velocity at
13 4mM (V₄) and the final EMG levels. **RESULTS:** Final EMG levels for the intact leg had a
14 very strong relationship with vLT ($r=0.77$, $p=0.001$) and a strong relationship with V₄
15 ($r=0.68$, $p=0.008$). Final EMG levels for the reconstructed leg had moderate relationship with
16 vLT ($r=0.47$, $p=0.09$) and V₄ ($r=0.52$, $p=0.06$). **CONCLUSION:** The neuromuscular
17 response of the intact leg during high intensity running shows strong to very strong
18 relationships with endurance markers. Failure of the ACLR leg to present relationships of
19 similar strength may indicate that chronic perturbations modify the ability of the local
20 muscular environment to tolerate sustained high intensity efforts.

21

22 Key words: EMG, lactate threshold, ACLR reconstruction, fatigue, running

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1 INTRODUCTION

2 The goal of the reconstruction of the anterior cruciate ligament in competitive athletes is
3 to achieve return to pre-injury exercise levels. In this regard, optimal muscle activation is of
4 great importance, especially during sustained athletic efforts. Previous research indicated that
5 muscle activation levels (as assessed by surface EMG) do not differ between the operated and
6 the intact leg during activities of moderate intensity [1, 2, 3]. However, it has recently been
7 demonstrated that during high intensity exercise the operated leg exhibits a diminished EMG
8 response over time as compared to both the intact and a healthy control leg [4, 5]. These
9 studies demonstrated that during a 10-min high intensity running bout, both the intact and the
10 healthy control leg significantly increased EMG activity of the vastus lateralis by an average
11 of ~8% compared to the initial values. However, the operated leg did not increase EMG
12 activity of the vastus lateralis as compared to baseline.

13 From a physiological perspective, the most commonly used endurance markers that are
14 considered prerequisite for optimal performance during high intensity sustained efforts are the
15 maximal oxygen uptake (VO_{2max}), and various aspects of the lactate threshold (LT). Such
16 aspects are mathematically derived thresholds and fixed blood lactate thresholds (e.g the
17 4mM threshold) [6, 7, 8, 9, 10]. A number of previous studies have confirmed that these
18 endurance markers correlate positively with the local neuromuscular response measured as
19 change in EMG activity [11, 12, 13, 14, 15, 16, 17]. Thus, a higher velocity or power output
20 at lactate threshold is associated with increased EMG activity, indicating that during high
21 intensity exercise the local neuromuscular response is largely determined by physiological
22 factors [18, 19, 20].

23 However, since our previous research [4, 5] has identified that ACLR athletes display
24 different local neuromuscular responses, it is possible that in such individuals the
25 relationships between endurance markers and the local neuromuscular response are also

1 altered. Such potential alterations may have important implications for the rehabilitation as
2 well as the conditioning of ACL reconstructed athletes. Therefore the purpose of the present
3 study was to assess the relationships between selected endurance markers and the relative
4 increase in local EMG activity during high intensity running in ACLR athletes. Specifically
5 we hypothesized that in these athletes higher EMG levels will be related with higher values in
6 endurance markers and that the intact leg will display different relationships than the operated
7 leg.

8

9 **MATERIALS AND METHODS**

10 Fourteen amateur male soccer players [mean (SD) age, body weight and height, 24.8
11 (5.3) years, 77.3 (7.5) kg and 177 (5.3) cm] with ACL-reconstructed knees were recruited for
12 the present study. The operated athletes had undergone ACL reconstruction with bone-patella
13 tendon-bone (BPTB) graft, 18.5 (4.3) months before testing. ACL reconstruction was
14 performed sub-acutely within 6 months after the injury from the same surgeon. All operated
15 athletes had a unilateral ACL tear confirmed by MRI and arthroscopy. They all underwent the
16 same rehabilitation protocol, starting from the first post operative day with the use of passive
17 exercises. Return to sports was permitted 6 months after reconstruction according to the
18 following widely accepted criteria [21]: (1) Full range of motion, (2) KT-1000 side-to side
19 difference <3mm, (3) quadriceps strength >85% compared to the contralateral side, (4)
20 hamstrings strength 100% compared to the contralateral side, (5) hamstrings-to-quadriceps
21 strength ratio >70% and (6) functional testing >85% compared to the contralateral side. At the
22 time of data collection, no clinical evidence of knee pain and effusion was found in the ACL
23 reconstructed subjects. All subjects agreed with the testing protocol and signed a consent form
24 according to the Declaration of Helsinki. Ethical approval was granted from the Institutional
25 Review Board policies of our Medical School. Prior to any data collection, a clinical

1 evaluation was performed in all athletes by the same clinician. During this evaluation, the
2 Tegner and Lysholm scores were obtained, while anterior tibial translation was evaluated
3 using the KT-1000 knee arthrometer (MEDmetric Corp., San Diego, California) [22]. The
4 athletes reported to the laboratory having abstained from caffeine or food consumption for 4
5 hours and without vigorous training for 24h on two different occasions separated by 48 hours.
6 Experimental testing took place at an ambient temperature of 20 °C to 22 °C.

7 For their first visit to the laboratory, the athletes performed a GXT test to volitional
8 exhaustion to determine VO_{2max} , LT and the 4mM threshold [23]. During warm-up the
9 athletes performed 3 minutes walking at self-selected pace and 5 minutes jogging on a
10 treadmill (Technogym Runrace 1200, Italy) at a speed of $8 \text{ km}\cdot\text{h}^{-1}$ where heart rate and blood
11 lactate were measured. The incremental test begun at a speed of $10 \text{ km}\cdot\text{h}^{-1}$ with stage
12 increments of $2 \text{ km}\cdot\text{h}^{-1}$ every three minutes. During the test, expired gas samples were
13 measured using a breath-by-breath metabolic measurement system (CPX Ultima, Medical
14 Graphics, St Maul, MN, USA). This system uses a zirconium O_2 analyzer and an infrared CO_2
15 analyzer. The subjects wore a facemask and inspired-expired gas samples were collected
16 through a pneumotachograph (pre-Vent, Medical Graphics, St Maul, MN, USA) and analyzed
17 for O_2 and CO_2 . At the end of each stage, capillary blood samples were collected and
18 analyzed for lactate (Accutrend, Roche Diagnostics, Germany). Prior to each test, all
19 analyzers were calibrated according to the manufacturer's instructions. According to the
20 American College of Sports Medicine, criteria for VO_{2max} were (a) plateau in VO_2 (an
21 increase $< 2.1 \text{ ml}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$ despite an increase in running speed, (b) a respiratory exchange
22 ratio (RER) > 1.10 , (c) , and (d) maximal blood lactate values (LA_{max}) $> 8 \text{ mmol}\cdot\text{l}^{-1}$ [23]. In
23 all cases at least 2 out of 3 criteria were met. The velocity associated with LT (v_{LT}) was
24 determined according to the D_{max} method proposed by Cheng et al [24]. Velocity at the 4
25 mM fixed blood lactate threshold (V_4) was determined from the individual velocity-blood

1 lactate curves using linear interpolation [8, 9, 10]. The velocity that was used for the
2 subsequent high intensity running was set at 40% of the difference between $\text{VO}_{2\text{max}}$ and
3 lactate threshold ($v\text{D}$) and represents the upper limit of high intensity exercise [25]. All three
4 velocities were also expressed as % of $\text{VO}_{2\text{max}}$ [7, 8, 9, 25].

5 In the second visit to the laboratory, athletes were required to perform a 10-minute high
6 intensity run at the $v\text{D}$ velocity. During the 10 minute running bout, EMG data were collected
7 for 15 seconds at the 3rd and 10th minute. Gas exchange data were recorded simultaneously
8 breath-by-breath and heart rate was measured throughout the test. Blood lactate was measured
9 prior to running and immediately after termination of exercise. EMG traces were obtained
10 from the vastus lateralis muscle bilaterally using bipolar, circular, pre-amplified, pre-gelled
11 Ag/AgCl electrodes with 10 mm diameter and fixed inter-electrode spacing of 20 mm
12 (Noraxon Inc, Scottsdale, AZ, USA). It has been demonstrated that vastus lateralis and rectus
13 femoris show similar high relationships between EMG activity and lactate threshold during
14 cycling [12, 13, 14]. However, vastus lateralis was selected on the basis that shows higher
15 levels of activity compared to rectus femoris during the first part of stance phase of running
16 and acts as a shock absorber, thus protecting the cartilage and the graft from high impact
17 forces [26]. EMG data were recorded with a wireless 8-channel EMG system (Telemetry
18 2400T, Noraxon Inc, Scottsdale, AZ, USA) and displayed real-time on a personal computer
19 using dedicated software (MyoResearchXP, Noraxon Inc, Scottsdale, AZ, USA). The surface
20 of the skin was prepared by shaving hair, rubbing it with abrasive paper and cleaning it with
21 alcohol. The electrodes were fixed longitudinally over the muscle belly. Electrodes were
22 placed at the antero-lateral muscle bulge at $2/3$ of the proximo-distal thigh length [27, 28].
23 The visually largest area of muscle belly was selected using a contraction against manual
24 resistance. The ground electrode was placed on the lateral femoral condyle of the right leg

1 [27]. Electrodes and cables were secured with surgical tape, in order to avoid any interference
2 with the running pattern of the subjects.

3 Footswitches (Noraxon Inc, Scottsdale, AZ, USA) placed under the heel and big toes of
4 both legs were used to denote heel-strike and toe-off events. EMG was acquired at a sampling
5 rate of 1500 Hz. The raw EMG was measured in a band of 10 to 500 Hz, was full-wave
6 rectified, was high pass filtered (cut-off frequency at 20 Hz) with an 8th order Butterworth
7 filter to remove movement artifacts and was smoothed with a 100 ms RMS algorithm. Values
8 from 20 strides were averaged to calculate the mean peak amplitude during stance. A
9 preliminary review of the relevant literature revealed that EMG amplitude has been used in
10 many previous studies of similar design [11-13, 16, 17] and is considered to be a reliable
11 parameter to evidence EMG activity. Mean peak amplitude values from the 10th minute were
12 expressed as percentage of the initial values during the 3rd minute and were defined as the
13 final EMG values.

14 Statistical analysis: Data were expressed as means and standard deviation (mean \pm SD).
15 Pearson product moment correlations were used to determine the relationships between final
16 EMG values and the VO_2max (both as $\text{lt}\cdot\text{min}^{-1}$ and $\text{ml}\cdot\text{min}^{-1}\cdot\text{kg}^{-1}$), vLT, vLT expressed as
17 percentage of VO_2max (vLT %), V_4 , V_4 expressed as percentage of VO_2max (V_4 %), vD and
18 vD expressed as percentage of VO_2max (vD %). These relationships were identified for both
19 the ACLR and the intact leg.

20

21 **RESULTS**

22 Negative Lachman and pivot shift tests indicated that the static knee joint stability was
23 regained. The subjects had all returned to their previous level activity engaging running and
24 soccer specific training for 3-5 sessions/week. Lysholm score was 95 (94-96) and Tegner
25 score was 8 (range, 7-9). KT-1000 results revealed that the mean difference between the

1 anterior tibial translation of the reconstructed and intact sides was 1.6 mm (range, 1 to 2mm)
2 for the 134-N test and 1.8 mm (range, 1 to 2 mm) for the maximal manual test.

3 Mean values for the endurance markers are presented in Table 1. The vD high intensity
4 run was performed on average at 87.6% (4.4) of VO_2 max. Blood lactate values during the vD
5 run rose from an initial baseline value of 2.1 (0.3) mM to 7.6 (1.7) mM. All athletes
6 completed the 10 min vD run without being completely exhausted and indicated that they
7 could continue for “a few more minutes”.

8 **INSERT TABLE 1 ABOUT HERE**

9 Pearson moment product correlations and coefficients of determination between final
10 EMG activity of the intact ($EMG10_{int}$) and reconstructed leg ($EMG10_{rec}$) and each one of the
11 endurance markers are presented in Table 2. $EMG10_{int}$ demonstrated a very strong
12 relationship with vLT ($r=0.77$, $p=0.001$) and $V_4\%$ ($r=0.75$, $p=0.002$), a strong relationship
13 with V_4 ($r=0.68$, $p=0.008$) and vD% ($r=0.57$, $p=0.03$) and moderate relationships with vLT%
14 ($r=0.46$, $p=0.09$) and vD ($r=0.44$, $p=0.11$). Finally, $EMG10_{int}$ displayed only weak relationship
15 with VO_2 max expressed either as absolute ($r=-0.19$, $p=0.51$) or relative values ($r=-0.18$,
16 $p=0.54$). $EMG10_{rec}$ demonstrated moderate relationships with vLT ($r=0.47$, $p=0.09$), V_4
17 ($r=0.52$, $p=0.06$) and $V_4\%$ ($r=0.5$, $p=0.07$), weak relationships with vD ($r=0.2$, $p=0.49$) and
18 vD% ($r=0.24$, $p=0.42$) and very weak relationships with vLT% ($r=0.08$, $p=0.87$) and VO_2 max
19 expressed as either absolute ($r=0.05$, $p=0.87$) or relative ($r=-0.03$, $p=0.92$) values.

20 **TABLE 2 ABOUT HERE**

21

22 **DISCUSSION**

23 The purpose of the present study was to assess the relationships between endurance
24 markers and the relative increase in local EMG activity during high intensity running in
25 ACLR athletes. Specifically we hypothesized that in these athletes higher EMG levels will be

1 related with higher values in the endurance markers and that the intact leg will display
2 different relationships than the operated leg.

3 The major finding of the present study was that EMG activity of the intact leg during
4 high intensity running demonstrated very strong relationships with vLT , V_4 % and strong
5 relationships with V_4 and vD %, while EMG activity of the operated leg showed only
6 moderate relationships with vLT , V_4 and V_4 %. The strong to very strong relationships
7 ($r=0.57-0.77$) established for the intact leg are in line with previous studies that have
8 suggested a strong physiological link between myoelectric changes at fatigue and the lactate
9 threshold [15, 18, 19, 20].

10 During sustained fatiguing running performed at the vD velocity which averaged 87.6%
11 (4.4) of VO_{2max} , EMG of the working muscles tends to increase over time as a result of (i)
12 increased firing rate (rate-coding) of the already activated muscle fibers, (ii) recruitment of
13 previously inactive fibers, especially the type II and (iii) both of above mechanisms [18, 29,
14 30]. On the other hand at these high exercise intensities there is substantial accumulation of
15 metabolic by-products such as lactic acid, H^+ , P_i , which may lead to drop out of mechanically
16 failing muscle fibers and decreased EMG activity over time [31]. Thus, final EMG levels
17 during sustained high intensity running depend upon the balance between these two opposing
18 trends. Furthermore, given the fact that lactate threshold represent the balance between
19 accumulation and clearance of metabolic by-products in the exercising muscles [32, 33], it is
20 not surprising that EMG activity of the intact VL showed strong to very strong relationships
21 with the two markers indicative of the lactate threshold, namely vLT and V_4 . We hypothesize
22 that the athletes that had higher vLT and V_4 values were able to increase their EMG activity
23 of the intact leg to higher levels probably due to the fact that were better able to sustain the
24 accumulation of metabolic by-products which tends to suppress EMG amplitude.

1 We were able to demonstrate only a weak relationship between EMG activity of the
2 intact leg and VO_2max . This is line with Bassett and Howley [6] who concluded that VO_2max
3 is mainly linked to central adaptations, presumably maximal cardiac output, while lactate
4 threshold incorporates both central and peripheral adaptations [7, 34]. Thus, lactate threshold
5 may more closely reflect the ability of the local muscle environment to tolerate fatiguing
6 exercise without compromising myoelectrical activity.

7 Our results further indicated that the EMG activity of the reconstructed leg showed only
8 moderate relationships with $v\text{LT}$, V_4 and $V_4\%$. Several explanations can be given for the lack
9 of a strong relationship between EMG activity in the reconstructed leg and the endurance
10 markers. Recent studies have presented evidence supporting the notion that selective fiber II
11 atrophy may occur in the involved quadriceps after ACL reconstruction [35, 36, 37].
12 Furthermore, alterations in activity patterns following surgery and subsequent retraining may
13 alter motor unit activation [38]. In addition, loss of joint afferent information may lead to
14 quadriceps weakness and subsequent selective hypotrophy of type II muscle fibers [39]. Thus,
15 the reconstructed leg may display a decreased potential for progressive recruitment and this
16 would only be apparent when performing at high intensities [30]. This seems plausible since it
17 has been suggested that in the case of large muscles, additional recruitment and not increases
18 of firing rate is the major strategy used to generate the required forces over a wide range of
19 submaximal exercise intensities [40]. The above hypothesis is also supported by the fact that
20 the reconstructed leg does not increase EMG activity over time during sustained high
21 intensity exercise [4, 5]. On the other hand, greater suppression of the EMG signal of the
22 reconstructed leg due to higher accumulation of metabolic by-products as compared to the
23 intact leg, may also explain the lack of relationship between EMG activity and endurance
24 markers and the lack of increase in EMG activity over time [31]. This hypothesis is supported
25 by the fact that long-term muscle detraining in endurance athletes, such as soccer players

1 undergoing knee surgery, induces a transformation of Ila fibers (fast-oxidative, fatigue
2 resistant) to Iib fibers (fast-glycolytic, fatiguable) [41, 42]. Thus loss of the aerobic metabolic
3 characteristics of Ila fibers of the operated leg may suggest that when performing at high
4 intensities (above the lactate threshold) these fibers are subjected to greater metabolic
5 disturbances compared to the “normal” Ila fibers of the contralateral leg. In any case, the
6 above two hypotheses imply that high intensity exercise is not tolerated in the same manner
7 by the VL of the intact and the reconstructed leg and that fatiguing exercise may provoke
8 greater physiological strain on the VL muscle of the reconstructed leg compared to the VL
9 muscle of the intact leg.

10 Given the interdependence between neuromuscular and metabolic systems, an important
11 implication of the present study is the potential for improved neuromuscular response of the
12 reconstructed leg following endurance training. Usually, improvement of aerobic fitness of
13 athletes following ACL reconstruction is performed after the athlete is discharged from
14 formal rehabilitation and only in the case of elite athletes during accelerated rehabilitation
15 [43]. It should be anticipated that an aerobic endurance training program aiming at peripheral
16 (muscle) adaptations would improve neuromuscular response by improving lactate tolerance
17 and accumulation of metabolites implicated in the fatigue process during high intensity
18 exercise [7, 8, 29, 34, 44]. To the best of our knowledge this is the first study that
19 investigated directly the relationship between the local neuromuscular response and
20 endurance markers in ACL reconstructed athletes. Previous studies on healthy subjects have
21 assessed extensively the relationship between the local neuromuscular response and
22 endurance markers during high intensity cycling [11, 12, 13, 14, 16, 17]. Our approach
23 enabled us to extend our findings to intense running which represents a highly functional
24 activity for the ACL reconstructed athlete. Furthermore, using preliminary VO_{2max} and

1 lactate threshold testing we assigned high intensity exercise according to individual fitness
2 levels. Thus, all subjects exercised under identical controlled conditions.

3 In the present study only ACL reconstructed athletes with bone-patellar tendon-bone
4 autograft were recruited. Thus, it is unknown if a similar response pattern will be observed in
5 athletes with a different graft such as hamstrings. Furthermore it is accepted that
6 proprioceptive deficits occur in most patients after ACL rupture and that persist to some
7 degree after ACL reconstruction [45]. However we believe that this should have a minimal
8 effect in our study because, a) our players were all treated with the same rehabilitative
9 protocol, which emphasized on proprioceptive re-training and b) our study design involved
10 treadmill running which is a cyclic highly repeatable two-dimensional motion. It should also
11 be acknowledged that EMG recordings should be performed with great care and the results
12 should be interpreted with caution when it comes to dynamic muscle contractions and
13 especially whole body exercises such as running. With that in mind, signal capturing,
14 recording and processing were performed according to established guidelines [27, 28]. We
15 selected a fixed epoch for the period of contraction in our study. Thus, we examined electrical
16 activity developed solely during the stance period, thereby reducing to some extent the role of
17 the signal non-stationarities with respect to the other effects being studied [46]. Furthermore,
18 the peak activity of many (successive) steps was averaged providing a reasonable estimation
19 of peak electrical activity during every recording period and minimizing within subject
20 variability. In addition, our study design involved repeated measures, that is, the final value of
21 the EMG activity was normalized to its original value while the electrodes remained attached
22 during the whole task, thereby overcoming the between-subject variability of EMG amplitude
23 [47].

24

25

1 **CONCLUSIONS**

2 In conclusion, the neuromuscular response of the intact leg during high intensity
3 exercise shows strong to very strong relationships with endurance markers that are indicative
4 of local muscle adaptations to endurance training. Failure of the ACLR leg to present a
5 similar response may indicate that chronic perturbations may impair the normal physiological
6 response and/or that variables other than endurance markers may also be implicated in the
7 ability of the local muscle environment to tolerate sustained high intensity efforts.

8

9

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13

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21 **TABLE LEGENDS**

22 **TABLE 1.** Mean group values for the endurance markers. Values are mean \pm 1 standard
23 deviation.

24 **TABLE 2.** Correlation coefficients (r) and coefficients of determination (r^2) between
25 EMG activity and endurance markers.