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Functional and morphological changes in the quadriceps muscle induced by eccentric training after ACL reconstruction

Alterações funcionais e morfológicas do quadríceps induzidas pelo treinamento excêntrico após reconstrução do LCA

Jamilson S. Brasileiro¹, Olga M. S. F. Pinto², Mariana A. Ávila², Tania F. Salvini²

Abstract

Objectives: The purpose of this study was to investigate the contributions of functional and morphological factors in the recovery of the quadriceps muscle after anterior cruciate ligament (ACL) reconstruction. **Methods:** Nine subjects (31.3±5.8 years) underwent eccentric exercise sessions twice a week for 12 weeks. Quadriceps muscle function was evaluated using an isokinetic dynamometer (isometric and eccentric peak torque) and electromyography (RMS). Morphological changes were measured using magnetic resonance imaging. **Results:** The initial evaluation showed a significant deficit in knee extensor torque in the involved limb and significant muscle atrophy along the length of the quadriceps. EMG activity was lower in all tested situations. Eccentric training significantly increased isokinetic torque (from 199±51 to 240±63, $p<0.05$, respectively) and quadriceps area, with the greatest hypertrophy in the proximal thigh region (from 169±27 to 189±25.8 cm², $p<0.01$). The EMG activity of vastus medialis increased after the first six weeks of eccentric training. The increased extensor torque was correlated with quadriceps cross-sectional area ($r=0.81$, $p<0.01$) and EMG activity ($r=0.69$, $p<0.05$). After twelve weeks of training, there was a correlation only between torque and cross-sectional area ($r=0.78$, $p<0.01$). **Conclusions:** 1) eccentric training proved to be a potent resource for the quadriceps recovery, both morphologically and functionally, 2) the contributions of functional and morphological factors varied according to the length of training.

Key words: anterior cruciate ligament; electromyography; magnetic resonance; quadriceps muscle.

Resumo

Objetivos: O propósito deste estudo foi avaliar as contribuições dos fatores funcionais e morfológicos na recuperação da força muscular do quadríceps femoral após reconstrução do Ligamento Cruzado Anterior (LCA). **Métodos:** Nove indivíduos (31,3±5,8 anos) foram treinados por meio de contrações excêntricas máximas, duas vezes por semana, durante 12 semanas. A função do quadríceps foi avaliada pela dinamometria isocinética (pico de torque isométrico e excêntrico) e pela eletromiografia (EMG). As alterações morfológicas foram mensuradas por meio de ressonância magnética (RNM). Na avaliação inicial, observou-se significativo déficit no torque extensor do joelho do membro acometido, com hipotrofia muscular de todo o quadríceps e redução na atividade EMG, quando comparado ao membro não-acometido. **Resultados:** O treinamento excêntrico produziu aumento no torque excêntrico a 30°/s (de 199±51 Nm para 240±63 Nm, $p<0,05$) e no volume muscular, sendo que maiores hipertrofias ocorreram na região proximal da coxa (de 169±27 para 189±25,8 cm², $p<0,01$). A atividade EMG do Vasto Medial (VM) aumentou nas primeiras seis semanas de treinamento. O aumento no torque extensor demonstrou correlação positiva com o aumento no volume ($r=0,81$, $p<0,01$) e na atividade eletromiográfica (EMG) ($r=0,69$, $p<0,05$) nas primeiras seis semanas. Após 12 semanas de treinamento, houve correlação apenas entre o aumento do torque e do volume ($r=0,78$, $p<0,01$). **Conclusões:** 1) O treinamento excêntrico mostrou-se como potente recurso tanto na recuperação dos fatores morfológicos como funcionais do músculo quadríceps; 2) A contribuição dos fatores neurais e morfológicos varia em função do período de treinamento.

Palavras-chave: ligamento cruzado anterior; eletromiografia; ressonância magnética; quadríceps femoral.

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¹ Physical Therapy Department, Universidade Federal do Rio Grande do Norte (UFRN), Natal, RN, Brazil

² Physical Therapy Department, Universidade Federal de São Carlos (UFSCar), São Carlos, SP, Brazil

Correspondence to: Jamilson Simões Brasileiro, Universidade Federal do Rio Grande do Norte, Campus Universitário, Departamento de Fisioterapia, Av. Senador Salgado Filho, 3000, Lagoa Nova, CEP 59.078-970, Natal, RN, Brasil, e-mail: brasileiro@ufrnet.br

Introduction ::::

The anterior cruciate ligament (ACL) is one of the most frequently injured ligaments¹. Several studies have demonstrated that even after ACL reconstruction patients are discharged from rehabilitation, significant deficits in quadriceps muscle strength and cross-sectional area still remain. These deficits can persist for months or even years²⁻⁷. It has been shown that the recovery of knee extensor torque is an essential element for functional rehabilitation of the lower limb after reconstruction since the return to functional activity is strongly correlated with the ability of the quadriceps femoris to generate force^{3,5,8}.

Understanding the physiological basis of muscle strength recovery in patients submitted to ACL reconstruction is fundamental for developing an appropriate rehabilitation program. Without correct identification of the factors involved in the loss and recovery of strength, it is difficult to select the most adequate physical therapy resources to use.

Neural and morphological participation have been suggested as the two main factors in the recovery of quadriceps strength. Neural factors are largely related to better motor unit efficacy during muscle contraction. It is known that the greater the number of motor units activated, the greater the resulting strength. Moreover, better timing of muscle fiber activation associated with an increase in triggering rate optimizes the excitation-contraction coupling which leads to further elevation of muscle force production⁹⁻¹³. It has been observed that in post-traumatic situations, inhibitory mechanisms act to reduce neuromuscular excitement, which leads to a reduction in muscle contractile capacity⁹. This activity can be measured by the RMS (*Root Mean Square*) of the electromyographic signal¹⁴.

On the other hand, morphological factors generally refer to the muscle cross-sectional area. The larger the fiber diameter, the greater the number of cross-bridges built and, thus, the higher the capacity to generate force^{9,11}. Reduced trophism is often associated with traumatic injuries and can be measured by imaging techniques such as ultrasound, computerized tomography or, preferably, magnetic resonance imaging (MRI). Isokinetic dynamometry can reveal changes in neuromuscular function, such as those related to peak torque, average torque, power, total work, time and peak torque angle. In current practice, it is an important tool for functional assessment due to its high level of reliability and validity.

It has been suggested that initial strength gains would result from the contribution of neural factors and that later phase gains would be due to muscular hypertrophy^{9,11,13}. However, studies evaluating these factors have been restricted to healthy subjects. Studies involving subjects submitted to this kind of rehabilitation are scarce. Lieber¹³ has called attention to the lack of studies with rehabilitation subjects, suggesting that if strength deficits occur principally due to neural factors, rehabilitation should focus,

above all, on resources that stimulate neural activation, such as neuromuscular electrical stimulation and the use of stretch reflexes and balance reactions. However if deficits are due to morphological factors such as hypertrophy, he states that the treatment focus should be directed to maximal resistance exercise.

The adequate identification of the factors responsible for strength deficit in this population will allow the physical therapist to select the best resource for each phase of the rehabilitation process. For this reason, the present study investigated the contribution of neurological and morphological factors to the recovery of quadriceps femoris function in patients submitted to ACL reconstruction.

Methods ::::

Subjects

Nine sedentary male subjects (31.3±5.8 years-old) who had been submitted to unilateral ACL reconstruction involving the middle third of the patellar tendon participated in this study. All the procedures were performed by the same surgeon. The subjects were between the 9 and 10 months post-surgery (mean of 9.4±0.7 months), which can be considered in the late rehabilitation phase. All the subjects were submitted to the same rehabilitation protocol, which began immediately after surgery. Those with other associated injuries, a previous history of trauma in the contralateral knee or joint pain during training were excluded from the study.

All the volunteers were informed about the objectives of this study and gave written informed consent before participating. This study was approved by the Research Ethics Committee of the Universidade Federal de São Carlos (UFSCar), São Carlos, SP, Brazil under protocol number 013/2004.

Procedures ::::

Analysis of the cross-sectional area of the quadriceps (CSA)

To obtain the images of the quadriceps femoris, a Torm[®] 0.5 Tesla MRI scanner (São Carlos, SP, Brazil) was used with the subject in the supine position. The muscle image acquisition parameters were as follows: a) field intensity = 0.5 tesla; b) time of repetition (TR) = 430 ms; time to echo (TE) = 26 ms; d) acquisition matrix = 256 x 192; e) slice thickness = 8 mm; f) slice gap = 0 mm; g) radio frequency bandwidth = 16 KHz.

The images were measured every 3.2 cm¹⁵ with *Axionvision* v3.0 software (*Carl Zeiss*[®], Germany).

In each evaluated plane, the muscle area was measured consecutively three times by the same rater and the arithmetic mean of these three measures was considered as the CSA of the quadriceps (in cm²). Previous analysis revealed an intra-rater reliability level above 0.95.

Isokinetic dynamometry

The subjects initiated the isokinetic tests after a 5 min warmup on a stationary cycle ergometer with 25 W resistance at 20 Km/h. Then they performed passive self-stretching of the quadriceps femoris in both limbs. The stretching maneuver consisted of maintaining the knee in complete flexion with the hip extended to the maximal tolerable amplitude while in the orthostatic position. Three 30 s repetitions were performed with a 30 s interval between each repetition.

An isokinetic dynamometer (*Biodex[®] Multi-Joint System 3*, Biodex Biomedical System Inc., New York) was used for isometric and isokinetic torque assessment as well as for training. Subjects were seated in the chair, which was inclined 5° backwards from vertical, with their trunks stabilized by two belts while the resistance arm was positioned at the distal portion of the leg. The mechanical axis of the dynamometer was aligned with the lateral epicondyle of the femur. Adjustments to correct for the effects of gravity on torque were made with the knee at 60° of flexion and calculated with the dynamometer's software, as recommended by Dvir¹⁶. The assessment was initially performed with the non-injured limb and then with the injured one.

After a short period of familiarization with the equipment, the subject performed three 5 s maximal isometric voluntary contractions (MIVC) with the knee at 60° of flexion, resting for 2 min between each contraction. The extensor peak torque between the three trials was registered for further analysis.

The isokinetic evaluation was initiated after an interval of three minutes, with the dynamometer adjusted to eccentric mode and with a speed of 30 and 120°/s. The subject's knee was passively extended to 20° (considering zero as a complete extension) and then the subject was asked to perform a full extension until reaching a 90° angle, totaling an active range of 70°. Each subject performed five repetitions at 30°/s and five repetitions at 120°/s. During the execution of maximal voluntary contractions, verbal encouragement was given by the same evaluators.

Electromyographic activity (EMG)

The EMG measurement procedure was as follows: the surface of the skin was shaved and cleaned with alcohol. The receiving electrodes (Lynx Tecnologia Eletrônica[®], Brazil) were then positioned over the rectus femoris (RF), vastus lateralis (VL) and vastus medialis obliquus (VMO) muscles according to recommendations of

SENIAM (*Surface Electromyography for Non-Invasive Assessment of Muscles*), with the reference electrode fixed at the tibial tuberosity.

EMG activity was captured simultaneously with the torque records by means of a 12 bit analog-to-digital converter (CAD, 12/36-60K – Lynx Tecnologia Eletrônica[®]) with a 16-channel signal conditioner module (MCS 1000). Signals were captured with *Aqdados*, v5.0 (Lynx Tecnologia Eletrônica, Brazil), sampled at 1 KHz and filtered (20 to 500 Hz) according to DeLuca¹⁷. The electrode had a 20x internal gain and a common mode rejection rate of over 80 dB. Since the gain programmed in the analog-to-digital converter was 50x, the signal was amplified 1000 times.

The obtained data was stored and subsequently converted to *ASCii* format. After processing, the files were analyzed in *MatLab[®]* v5.0, by means of which the RMS value was identified for EMG signal analysis.

The initial evaluation procedures were repeated after six weeks of training (evaluation 2) and again after 12 weeks of training (evaluation 3).

Training protocol

Each subject was submitted to a maximal eccentric exercise training session twice a week for 12 weeks. Each session began with a short warmup followed by stretching like that performed before the evaluations.

The subject was then positioned in the isokinetic dynamometer as described above. The same procedures used in the isokinetic evaluation were used for eccentric training, with three sets of ten repetitions performed. Only the injured limb was trained and only at the speed of 30°/s.

Statistical analysis

For the analysis and interpretation of the results, *Statistical Package for Social Sciences* (SPSS) v15.0 was used. The normality of all data was initially verified with Kolmogorov-Smirnov. The peak torque analysis, EMG activity and CSA obtained in the three evaluations were submitted to a new repeated-measures ANOVA for comparison. For all significant causes of variation, *post-hoc* Tukey was used. Pearson's Correlation was used to determine the relation between the dynamometric, EMG and CSA data. The significance level was set at 5% for all tests.

Results

Peak torque (PT)

The highest PT was found in the eccentric contraction at 30°/s, followed by the eccentric contraction at 120°/s. The

lowest PT was registered during the isometric contractions (Figure 1).

In the eccentric evaluation at 30°/s, the training had already produced an increase in PT by the sixth week (from 227±56 to 254±65 Nm, $p=0.031$) that continued until the end of the training (290±64 Nm, $p=0.007$). Significant increases in PT were also observed for isometric and eccentric contractions at 120°/s (velocity not used in training), which was verified only after 12 weeks of training (from 198±37 to 228±48 Nm, $p=0.041$ and from 200±51 to 240±63 Nm, $p=0.039$, respectively).

PT from the non-injured limb did not show significant variation over the course of the study.

Analysis of CSA

MRI images of the entire thigh showed that there was a significant increase in quadriceps CSA in the injured limb by the second evaluation that continued until the end of the study. In the non-injured limb, no alteration in the CSA of any of the measured regions was observed.

Individual analysis of each CSA level along the length of the thigh revealed that the trained limb presented non-homogeneous behavior with respect to recovery of quadriceps trophism (Figure 2).

No significant hypertrophy was observed in the distal portion of the thigh in the first six weeks of training (from 31.5±5.9 to 31.6±6.2 cm², $p=0.38$). By the end of the training period, an increase of 11.5% in measured area was observed (35.1±7 cm², $p=0.043$). The increase in this region was occurred mainly in the VM. In an intermediate region of the thigh, hypertrophy was observed by the end of the first six weeks of training (from 123±23 to 128±25 cm², $p=0.029$) and increased until the third evaluation (134±25 cm², $p=0.009$).

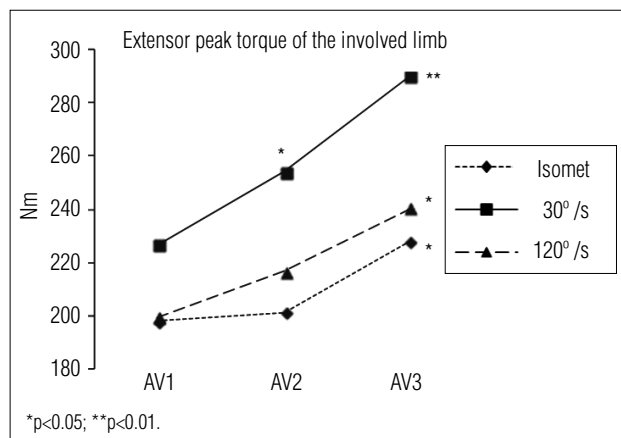


Figure 1. Isometric and eccentric (30 and 120°/s) knee extensor peak torque of the involved limb, pre-training (AV1), after six (AV2) and twelve weeks (AV3).

The studied region includes the vastus intermedius (VI), vastus medialis (VM) and VL. In the proximal region of the thigh, hypertrophy was already observed after the six first weeks of training (from 166±26 to 175±28 cm², $p=0.009$), with a continuous increase until the 12th week (182±29 cm², $p=0.003$). In this region, are evidences the VI, VL and RF.

EMG activity

There was a significant increase in the RMS value of the VMO and VL only during the first six weeks of training (from 213±107 to 289±81 μV, $p=0.037$ and from 207±65 to 229±69 μV, $p=0.042$, respectively). The RMS and RF values did not undergo significant modifications during the training (Figure 3).

In contrast, the statistical analysis did not reveal significant differences between the EMG values registered during the isometric and eccentric contractions at 30°/s for the same muscle portion ($p>0.05$).

Correlations between peak torque, cross-sectional area and EMG activity

In the first six weeks of training, significant increases in isokinetic PT at 30°/s were observed, followed by an increase in the average EMG activity of the three analyzed vastus muscles and a significant increase in average CSA along the length

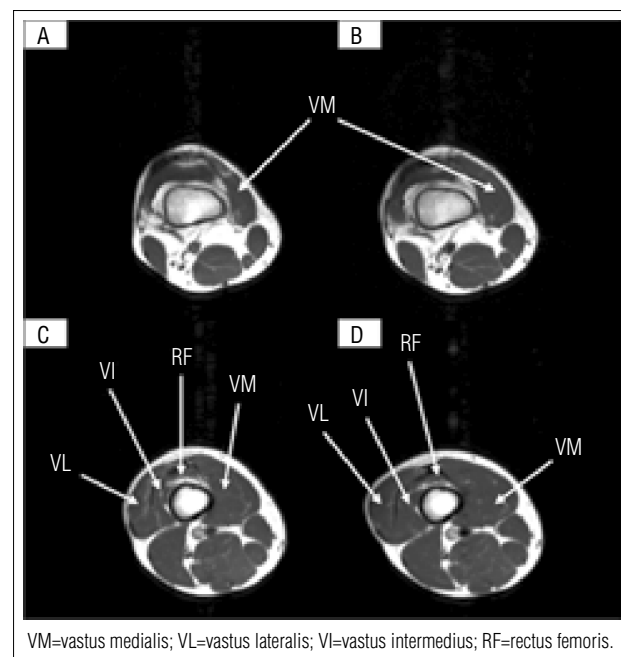


Figure 2. Representative axial T2-weighted MRI of the involved limb, before (A and C) and after training (B and D) of the same subject. The images were obtained between 6 cm (A and B) and 20 cm (C and D) from the proximal border of the patella.

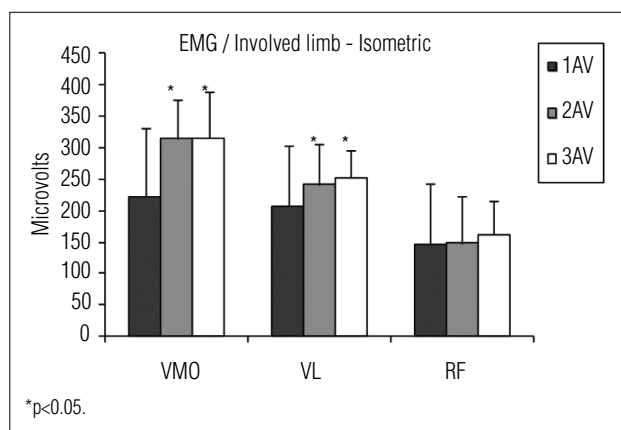


Figure 3. Electromyographic activity (RMS) of the VMO, VL and RF during isometric contractions of the involved limbs, pre-training (AV1), after six (AV2) e twelve weeks (AV3).

of the thigh. There was a strong correlation between the variables PT and CSA ($r=0.81$, $p=0.020$) and a moderate correlation between the variables PT and EMG ($r=0.69$, $p=0.037$).

In the second six weeks, PT also varied significantly from the values observed in the second evaluation. This gain was followed by an increase in muscle CSA, but no change in EMG activity was observed. In this phase, a strong correlation was also maintained between PT and CSA ($r=0.78$, $p=0.01$), although no correlation was observed between PT and EMG ($r=0.28$, $p=0.13$).

Discussion

The purpose of the present study was to analyze the contribution of neural and morphological factors to the recovery of quadriceps strength after a training period involving maximal eccentric contractions.

At the end of the training, although torque gains were significant, a residual deficit of 18 to 29% persisted between the injured and non-injured limbs. Hiemstra et al.⁵ found a global deficit of 25.5% in extensor torque when evaluating 24 subjects submitted to ACL reconstruction after one year of recovery. The same authors emphasized the scarcity of studies involving eccentric evaluations. This is important since, theoretically, greater deficits could be revealed in this form of contraction, since greater muscle tension is generated.

The existence of such a torque deficit may reveal a change in recruitment pattern, mechanical joint changes or modifications in muscle properties due to physical deconditioning. Urbach et al.¹⁸ found torque deficits without the presence of significant changes in neural recruitment in patients like those evaluated in the present study, which suggests that, in addition

to possible deficits of voluntary activation, disuse atrophy may still be present.

The proposed eccentric training program was efficacious in increasing muscle trophism, with gains that appeared after six weeks and gradually increased until the end of the 12th week.

Some studies have suggested that maximal eccentric training is more efficient for increasing muscle trophism than concentric treatment¹⁹⁻²¹. Since more strength can be generated eccentrically, this training modality would produce more overload in the muscle, which would induce higher protein synthesis^{22,23}. It has also been suggested that this would occur due to a greater recruitment of type II fibers during the contractions^{10,24,25}, since these fibers typically demonstrate greater potential for hypertrophy than type I fibers^{13,19,20}.

Lieber¹³ and Enoka²⁴ have suggested that few muscle trophism changes can be observed during the initial weeks of training and that the recorded torque gains are almost exclusively attributed to neural changes. However, our study demonstrated a significant increase in CSA in the first six weeks of training, which suggests that the process of hypertrophy in these subjects may be different from that observed in healthy subjects.

Another result observed in our study was the difference in CSA increase between the proximal and distal extremes of the quadriceps. The proximal region presented significant hypertrophy after the first six weeks of training, whereas no gains were registered in the distal region.

Significant changes in hypertrophy between different parts of the quadriceps have been demonstrated in other studies^{10,26}. One possible justification for this would be the difference in the proportion of type I and II fibers between the different beams of the quadriceps muscle group²⁷, bearing in mind that type II fibers have the greatest potential for hypertrophy^{13,19,20}. Narici et al.²⁶ not only found significant differences among portions, but also within the same portion. In their study, the greatest gains were found in the RF (27.9%), followed by the VL (19.5%), VM (18.7%) and VI (17.4%). These authors also observed that a parallel increase in the torque and CSA of the quadriceps only occurred after the second month of training.

The proposed training program was also effective in increasing the amplitude of the EMG signal in the trained limb. Significant increases in RMS values were recorded in VMO and VL during the first six weeks of training. After this period, the values remained stable until the end of the study. There was no significant change in the RF RMS value throughout the training period.

Several studies have demonstrated an increase in EMG amplitude after periods of training, suggesting that in response to exercise there is a correspondent increase in neural discharge in the muscle fibers^{24,28}. Data from the present study show increased RMS values after training, both in isometric and eccentric

evaluations. Any apparent quadriceps neural dysfunction seems to be restored in the initial phases of training.

It has been proposed that neural factors would have greater importance in strength development during the initial stages of training and that the gradual subsequent increase in hypertrophy would gain influence until becoming the main factor responsible for changes in muscle strength^{9,11-13,25}. The present study evaluated both factors, and our results diverge from those of some authors.

The majority of studies do not show significant gains in muscular trophism in the initial phases of training. Hortobágyi et al.²⁵ observed that the initial adaptations to resistance training are almost exclusively neural. MacDougall et al.²⁹ found increases in muscle strength before any measurable sign of hypertrophy could be observed. Enoka²⁴ suggests that significant increases in cross-sectional area were not apparent before the eighth week of training.

However, one factor that could explain these divergent results is that all of the above-cited studies used healthy subjects. It is possible that the mechanisms involved in strength increases in non-injured limbs are different from those found after a period of disuse that includes neural inhibition and atrophy. Lieber¹³ has pointed out the "urgent necessity" of this type of study in patients submitted to rehabilitation programs. If strength recovery is primarily due to neural factors, the treatment should emphasize mechanisms of neuromuscular activation. If, however, recovery is mainly due to morphological factors, the emphasis should be directed to muscle strengthening exercises.

Our data show that in the initial training phase both neural and trophic factors contributed to torque increase. However, in the second half of the training period, only hypertrophy mechanisms influenced recovery. Thus, our data suggest that, in the

initial phases of muscle strength training programs, strength recovery would result from a combination of factors, involving increases in muscle cross-sectional area and contractile capacity. During this period, resources emphasizing neuromuscular activation (such as the use of electrical stimulation, stretch reflexes and balance reactions) should be associated with resources for increasing cross-sectional muscle area (such as maximal eccentric exercises). In the posterior phase, training should emphasize trophic factors, focusing on counter-resisted exercises. These factors should be considered when prescribing rehabilitation programs.

Conclusion ::::

The results of the present study suggest that the increased knee extensor torque of patients submitted to ACL reconstruction is due to an initial association of neural and morphological factors, while trophic changes are predominant in later stages. The study also showed that maximal eccentric exercises are a powerful kinetic therapy resource, facilitating both muscle strength and trophic recovery. It should be pointed out that it is necessary to consider the level of transplant maturation when applying a rehabilitation program after an ACL reconstruction. In other post-traumatic situations, factors such as pain, swelling and joint effusion must also be taken into account.

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