Title: The progression of isokinetic knee strength following matrix-induced autologous chondrocyte implantation: implications for rehabilitation and return to activity.

Running Title: Knee strength after MACI.

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ABSTRACT

Context: Matrix-induced autologous chondrocyte implantation (MACI) is an established technique for the repair of knee chondral defects. Despite the reported clinical improvement in knee pain and symptoms, little is known on the recovery of knee strength and its return to an appropriate level compared with the unaffected limb.

Objective: To investigate the progression of isokinetic knee strength and limb symmetry following MACI.

Design: Prospective cohort.

Setting: Private functional rehabilitation facility.

Patients: 58 patients treated with MACI for full-thickness cartilage defects to the femoral condyles.

Intervention: MACI and a standardized rehabilitation protocol.

Main Outcome Measures: Pre-operatively and at 1, 2 and 5 years post-surgery, patients underwent a three-repetition maximum straight leg raise (3RM-SLR) test, as well as assessment of isokinetic knee flexor and extensor torque and hamstring/quadriceps (H/Q) ratios. Correlation analysis investigated the association between strength and pain, demographics, defect and surgery characteristics. Linear regression analysis estimated differences in strength measures between the operated and non-operated limbs, as well as Limb Symmetry Indexes (LSI) over time.

Results: Peak knee extension torque improved significantly over time for both limbs, though was significantly lower on the operated limb pre-operatively and at 1, 2 and 5 years. A mean LSI of 77.0%, 83.0% and 86.5% was observed at 1, 2 and 5 years, respectively, while 53.4-72.4% of patients demonstrated an LSI ≤ 90% across the post-operative timeline. Peak knee flexion torque was significantly lower on the operated limb pre-operatively and at 1 year. H/Q ratios were significantly higher on the operated limb at all time points.

***Conclusions**:* While peak knee flexion and hip flexor strength were within normal limits, the majority of patients in this study still demonstrated an LSI for peak knee extensor strength ≤ 90%, even at 5 years. It is unknown how this prolonged knee extensor deficit may affect long-term graft outcome and risk of re-injury following their return to activity.

***Keywords**:* matrix-induced autologous chondrocyte implantation, rehabilitation, return to sport, strength.

INTRODUCTION

Matrix-induced autologous chondrocyte implantation (MACI) is an established technique for the repair of knee cartilage defects.[1-3] It is a two-stage procedure with an initial arthroscopic harvest of healthy cartilage, isolation and expansion of chondrocytes *ex-vivo*, and re-implantation of cells into the chondral defect. Cells are seeded directly onto a type I/III collagen membrane and fixed to the underlying subchondral bone with fibrin glue. Over time and, with the appropriate post-operative mechanical stimulus required for chondrocyte differentiation and development,[4-6] a durable load bearing tissue may be produced. A successful MACI outcome should return the patient to a pain-free and normally active lifestyle. However, despite the reported improvement in pain, symptoms and regeneration of hyaline-like articular cartilage after MACI,[1-3, 7-9] little has been reported on the recovery of knee strength, limb strength symmetry (or asymmetry) and any associated risk to the graft or rate of re-injury in the patients return to activity.

Thomee´ et al.[10] documented a number of factors that can affect the patients' successful return to sports after anterior cruciate ligament (ACL) reconstruction, including a lack of muscular strength and function. While evidence specifically for cartilage repair is scarce, one may assume that the recovery of knee strength is also of importance after MACI in order to provide ongoing knee joint support, as well as reduce the risk of graft and/or subsequent knee injury. In order to evaluate lower extremity function and neuromuscular performance, isokinetic dynamometry has been used extensively to identify deficits in knee flexor and extensor strength following several types of knee injury or surgery,[11-19] including MACI.[20-22] Though it may not mimic natural movements as is the case with dynamic functional testing, isokinetic dynamometry is generally accepted as a safe and valid method for assessing neuromuscular performance, training improvements and rehabilitation outcome.[12]

A frequently reported criterion for presenting the return of normal muscle strength and/or function is the Limb Symmetry Index (LSI).[21, 23-27] The LSI is the strength ratio of a specific muscle group, comparing the operated and non-operated limbs. The rationale of the LSI is to ensure that the injured and/or operated limb reaches an acceptable level in order to minimize the risk of overuse and/or further injury when returning to sport or strenuous work.[28] An LSI < 90% has been regarded as unsatisfactory for a variety of strength and functional tests, and may suggest that an individual is unsafe to return to regular sports activity.[10, 29, 30] Other researchers have considered an LSI below 85% as abnormal and not warranting the resumption of higher impact sports activities.[31, 32] While the association between LSI measures and re-injury risk has not been evaluated after any knee surgery to the best of our knowledge, the LSI does provide a valuable assessment of the return to an appropriate level of strength in the operated limb, compared with the non-operated limb. While LSIs have been reported extensively following ACL reconstruction for isokinetic strength assessment, they have not been specifically reported after MACI.

Thigh muscle atrophy and strength deficits have been outlined as a major problem in rehabilitation and the long-term evaluation of several types of knee surgeries, including chondrocyte implantation techniques.[12, 13, 15, 16, 20-22, 33-37] Reduced muscle atrophy and/or knee strength following MACI may be contributed to by the lengthy protective rehabilitation process required, a lack of adequate rehabilitation and/or, as reported following ACL reconstruction, a decrease in patient compliance over time during the rehabilitation process.[38] Furthermore, knee joint pain and effusion following injury and/or surgery can also act to reduce measurable knee strength,[12, 39] even in the absence of absolute strength deficits that may be attributable to relative limb disuse. Nevertheless, documentation of these LSIs in patients after MACI may provide insight into ongoing post-operative strength deficiencies, as well as pertinent information to the clinician in better rehabilitating these patients in their return to activity.

This study aimed to investigate the progression of post-operative isokinetic strength of the knee flexor and extensor muscles after MACI surgery. We hypothesized that: 1) a significantly lower peak knee flexor and extensor torque would be exhibited in the operated limb at 1 year post-surgery, compared with the non-operated limb, though no differences would be observed at 2 and 5 years, and 2) while a mean LSI $\leq$ 90% would be demonstrated at 1 year post-surgery, a mean LSI $\geq$ 90% would be observed at 2 and 5 years post-surgery.

METHODS

Design

Between June 2004 and November 2007, 68 patients underwent MACI to address full thickness medial or lateral femoral condylar defects to the knee, followed by a standardized rehabilitation program. Patients were assessed pre-operatively and at 1, 2 and 5 years post-surgery via a series of strength tests. These included a three-repetition maximum straight leg raise (3RM-SLR) test, as well as assessment of isokinetic knee flexor and extensor torque and hamstring/quadriceps (H/Q) ratios.

Patients

A total of 68 patients underwent MACI to address full thickness medial or lateral femoral condylar defects to the knee. For the current analysis, patients who underwent MACI on the contralateral limb within the assessment timeframe (n=2) were excluded, as were patients who did not undergo knee strength evaluation at 1, 2 and 5 years post-surgery (n=8). Therefore, this study analysis was undertaken in 58 patients (36 males, 22 females) with a mean age of 38.6 years (range 15-62), height of 1.74 m (range 1.55-1.91), weight of 81.3 kg (range 56.0-116.3), body mass index (BMI) of 26.7 (range 19.4-33.2) and defect size of 3.5 cm^2 (1.0-10.0). A mean of 1.2 prior procedures (range 0-4) had been undertaken prior to MACI on the involved knee, including: arthroscopy (n=35), partial meniscectomy (n=8), anterior cruciate ligament (ACL) reconstruction (n=5) and lateral release (n=2). A mean duration of symptoms of 9.3 years (range 1-46) was documented.

Patients were included if they were 15-65 years of age and deemed able to follow the rehabilitation program. Patients were excluded with a BMI > 35, had undergone prior total meniscectomy or had inflammatory arthritis on the operated limb, or had any reported deficit or prior injury/surgery history on the contralateral limb. Patients with ligamentous instability or varus/valgus abnormalities (> 3° tibiofemoral angle) were included, provided they were addressed prior to or at the time of MACI. Of the 58 patients in this study, no offloading osteotomies were required at the time of MACI, though concomitant surgical procedures that were undertaken included ACL reconstruction (n=1), posterior cruciate ligament (PCL) reconstruction (n=1) and partial meniscectomy (n=1). Patients provided written informed consent prior to study enrollment. Ethics approval was obtained from relevant university and hospital ethics committees, and research was undertaken according to the Declaration of Helsinki.

MACI Surgical Technique

MACI is a two-stage technique, where arthroscopic surgery was initially performed to harvest a sample of articular cartilage from a non weight bearing area of the knee. At this time the specific location, size and containment of the defect, ligamentous stability, and meniscus health were evaluated to determine the condition of the joint. After harvest, healthy chondrocytes were isolated, cultured for approximately four weeks (Genzyme, Perth, Western Australia) and seeded onto a type I/III collagen membrane (ACI-Maix, Matricel GmbH, Herzogenrath, Germany) three days prior to re-implantation. At the time of second-stage implantation, the chondral defect was prepared via an open medial or lateral mini-arthrotomy in a tourniquet-controlled field, by removing all damaged cartilage down to, but not through, subchondral bone. The resultant defect was measured and used to shape the membrane, secured to the bone using a thin layer of fibrin glue. Time was allowed for the glue to set and a further 2-3 minutes were given to allow the fibrin glue to cure before testing graft stability. Following assessment of graft stability the wound was closed.

Post-operative Rehabilitation

Immediate inpatient rehabilitation consisted of: continuous passive motion (0-30 degrees) within 12-24 hours after surgery; active dorsi- and plantar-flexion of the ankle to encourage lower extremity circulation; isometric contraction of the quadriceps, hamstrings, and gluteal musculature to maintain muscle tone;[40, 41] cryotherapy to control edema, and; teaching of proficient toe-touch ambulation through the affected limb. Following hospital discharge, and within two weeks of graft implantation, all patients began a coordinated out-patient rehabilitation program consisting of progressive exercise and graduated weight bearing over 12 weeks (Table 1), with ongoing education and advice on activity provided following this time.

Clinical Assessment

While clinical assessment was undertaken in all 58 patients at 1, 2 and 5 years post-surgery, it was undertaken in only 37 patients pre-surgery (prior to the initial arthroscopic biopsy required for cell harvest). Patients were referred upon booking of the arthroscopic cell harvest, though it was not forced upon them and completely up to the patient as to whether they made the appropriate pre-operative (pre-biopsy) visit for education and clinical assessment. Initially, a 3RM-SLR test was administered to assess the strength of the quadriceps and hip flexor musculature. The patient was assessed in a supine position, with the test leg fully extended, and the contralateral knee flexed at 90° with the foot flat. A weighted cable was attached to the ankle of the test leg, and the patient was instructed to lift the leg to the height of the contralateral bent knee. Every effort was made to minimize limb fatigue until a 3RM was attained. The initial weight was selected based on the size and estimated hip flexor strength of the individual patient, while the weight increment for each attempt was based on the reported and observed ease of the prior attempt. The patient was asked to attempt the first repetition and, if they felt they could attain three repetitions and desired a heavier weight, the test ceased and a heavier weight was attempted. The size of the weight lifted was continually increased until a 3RM was reached (recorded to the nearest 0.5 kg). The protocol was initiated on the non-operated limb and then repeated on the operated side, using the same starting weight and load increments undertaken on the non-operated limb.

Isokinetic strength of the quadriceps and hamstrings muscle groups was assessed using an isokinetic dynamometer (Isosport International, Gepps Cross, South Australia). Patients were seated in the dynamometer chair, so that the hips and knees were in a 90˚ position. The trunk and thigh were stabilized using rigid straps, and adjustments were made for thigh and leg length, in accordance with each patient's stature. Concentric knee extension and flexion strength was measured through a range of 0-90˚ of knee flexion, at a single isokinetic angular velocity of 90°/s. Each trial consisted of four repetitions: three low intensity repetitions of knee extension and flexion, immediately followed by one maximal test effort. Two trials on each lower limb were undertaken, alternating between the operated and non-operated limbs, with the first trial always undertaken on the non-operated side. Our choice of assessing the non-operated limb first was primarily to ensure the patient was fully aware of what was expected of them prior to maximally stressing their operated side, during a high knee stress activity that may not have been undertaken frequently by them. During each maximal effort, patients were asked to perform to their maximal muscle strength, while verbal encouragement was provided, standardized across all patients and trials. Patients were given adequate rest in between trials to minimize fatigue, though this was not standardized and based upon the patient's individual readiness to proceed. For all knee extension and flexion efforts, the peak torque value (Nm) and H/Q ratio were obtained. The H/Q ratio in this study was measured by dividing the peak concentric hamstrings torque by the peak concentric quadriceps torque.

The LSI was calculated for all strength measures by dividing the peak values on the operated limb by that recorded on the non-operated limb. A Visual Analogue Pain Scale (VAS) was employed to specifically determine the severity of knee pain during the isokinetic strength assessment at all time points.

Statistical Analysis

Correlation analysis between strength outcomes on the operated side and baseline patient demographics (age, height, weight, BMI), baseline defect parameters (size), injury/surgery history (prior procedures, duration of symptoms) and concomitant pain scores (VAS), was performed at pre-operative and 1, 2 and 5 year post-operative time points using Pearson's correlation coefficient.

The overarching aim of investigating the progression of strength measures in both limbs after MACI surgery was addressed using linear regression analysis with generalized estimating equations. The use of generalized estimating equations allows consideration of the non-independence of observations, i.e. for the correlation between repeated measures over time and side. Four separate regression analyses were performed, one for each strength measure (3RM-SLR, H/Q ratio, knee flexor and extensor peak torque). From each regression model, estimates were made of difference between the operated and non-operated limbs at all time-points, thus allowing the first research hypothesis to be addressed. In addition, changes in strength measures pre-operatively and at 1, 2 and 5 years post-surgery for each limb were also estimated. To address our second hypothesis, the mean LSI for knee flexor and extensor peak torque was calculated, and again linear regression analysis with generalized estimating equations was used to estimate the changes over time. LSI was also expressed as a percentage of patients with an LSI ≤ 85% and ≤ 90% at all time points. No correction was made for multiple testing, rather confidence intervals (95%) and associated p-values are provided for all differences, as is recommended practice in the field of biostatistics.[42] Models were evaluated for homogeneity of variance of residuals and absence of influential outliers by the examination of residual plots and standard linear regression diagnostics.

Regarding the missing baseline data for 21 patients at the pre-operative time-point, under this generalized estimating equation framework, the full sample is analyzed and estimates of means at each time point and side are made using information from the full sample at the other time points and the within-person correlation estimates. This ameliorates any potential bias in mean estimates which may be present in an available case analysis due to those with available data potentially being stronger or weaker. However, an available case analysis was also performed to allow an assessment of the impact of missing data on estimates. Statistical analysis was performed using Stata/IC 12.1 (StataCorp LP, TX USA), while statistical significance was determined at $p < 0.05$.

RESULTS

The 3RM-SLR, peak knee flexion and peak knee extension torque exhibited significant positive correlations with body weight and height (Table 2) at all time points. Age demonstrated a significant negative correlation with peak knee extension torque at all post-operative time points, as well as the 3RM-SLR at 1 and 5 years. The amount of prior knee procedures was only significantly correlated with peak knee extension torque pre-operatively, while the pre-operative duration of symptoms was significantly correlated with the 3RM-SLR at at all post-operative time points, as well as peak knee flexion torque at 2 and 5 years. The VAS during isokinetic testing was not significantly correlated with any of the pre- or post-operative strength measures (Table 2), with mean reported scores of 4.48 (range 0-10) pre-surgery, as well as 2.11 (range 0-9) at 1 year, 2.06 (range 0-8) at 2 years and 2.02 (range 0-7) at 5 years post-surgery.

Peak knee extension torque improved on both the operated and non-operated sides over the pre- and post-operative time line (Figure 1), with the largest and only statistically significant gains occurring between 1 and 2 years (Table 3 and Figure 1). Peak knee extension torque was significantly lower on the operated limb pre-operatively and at 1, 2 and 5 years post-surgery, compared with the non-operated limb (Table 3 and Figure 1). A mean LSI of 78.6% was demonstrated pre-operatively, while 77.0%, 83.0% and 86.5% was observed at 1, 2 and 5 years, respectively (Table 3 and Figure 1). Pre-surgery, 59.5% of patients demonstrated an LSI $\leq$ 90%, while 53.4-72.4% of patients demonstrated an LSI $\leq$ 90% across the post-operative assessment time points (Table 4). Furthermore, 48.3-70.7% of patients demonstrated an LSI $\leq$ 85% throughout the post-operative time line to five years (Table 4).

Peak knee flexion torque improved on both the operated and non-operated sides over the pre- and post-operative time line (Figure 1), with the largest and only statistically significant improvement occurring between 1 and 2 years post-surgery on the operated limb and between 2 and 5 years post-surgery on the non-operated limb (Table 5 and Figure 1). Peak knee flexion torque was significantly lower on the operated limb pre-operatively and at 1 year post-surgery, compared with the non-operated limb, though there were no differences at 2 and 5 years (Table 5 and Figure 1). A pre-operative mean LSI of 88.5%, and means of 90.5%, 99.9% and 98.2% were observed at 1, 2 and 5 years, respectively (Table 5 and Figure 1). While 48.6% of patients demonstrated an LSI ≤ 90% pre-surgery, 31.0-41.4% of patients demonstrated an LSI ≤ 90% across the post-operative assessment time points (Table 4).

The 3RM-SLR improved significantly on both the operated and non-operated sides through to 2 years post-surgery, though then decreased significantly between 2 and 5 years post-surgery on both limbs (Table 6 and Figure 1). There were no significant differences for the 3RM-SLR between the operated and non-operated limbs pre-operatively or at 1, 2 and 5 years post-surgery, with mean LSIs ranging from 96.7-100.8% (Table 6 and Figure 1). Pre-operatively, 18.9% of patients demonstrated an LSI ≤ 90%, while 17.2-27.6% of patients still demonstrated an LSI ≤ 90% across the post-operative assessment time points (Table 4).

There were no significant changes over the assessment timeline in the H/Q ratio for the operated or non-operated limbs, though the H/Q ratio was significantly higher on the operated limb at all post-operative time points, when compared with the non-operated limb (Table 7 and Figure 1).

The estimates reported in Tables 3, 5, 6 and 7 are the marginal mean estimates for the pre-operative time points, as 21 patients were missing strength measures pre-operatively. Therefore, the pre-operative to 1 year time difference and the pre-operative differences between the operated and non-operated limbs are estimated using information from the full sample at the other time points and the within-person correlation estimates. A sensitivity estimate using only observed data at these time points (n=37) showed very similar estimates with no change in statistical significance (Table 8), with the exception of the difference in the H/Q ratio between the operated and non-operated side pre-operatively, for which the effect size had narrower confidence intervals resulting in the p-value reducing from 0.065 to 0.002.

DISCUSSION

While MACI has become an established technique for the repair of knee chondral defects,[1-3] little has been reported on the recovery and symmetry of knee strength post-surgery. This study aimed to investigate the progression of post-operative isokinetic strength of the knee flexor and extensor muscles following MACI surgery to 5 years, comparing the operated and non-operated limbs.

At all pre- and post-operative time points, the peak knee extensor torque on the operated limb remained significantly lower than the non-operated limb. Pre-operatively, almost 60% of patients in this cohort demonstrated an LSI for knee extensor strength ≤ 85%, which appears similar to that reported previously in patients with articular cartilage defects.[21] Post-operatively, restoration of lower limb muscle function including isokinetic knee strength is considered important for a successful return to sports or physical activity.[10, 28, 43-45] While several LSI cut-offs have been reported in evaluating strength and functional performance,[24] both < 90%[10, 29, 30] and < 85%[31, 32] have been regarded as unsatisfactory, abnormal and may suggest that an individual is unsafe to return to regular sports activity. In the assessment of isokinetic knee extensor strength in this study, it would appear that the majority of patients in this cohort demonstrated an LSI ≤ 90% even at 5 years post-surgery, with 48.3% of patients still demonstrating an LSI for knee extensor strength ≤ 85% at 5 years. The majority of ACL-based studies only report LSI measures at the group level,[10, 16, 26, 31, 46-49] making it unclear as to whether ACL reconstruction sufficiently restores muscle function.[10] In this study, while the mean LSI for peak knee extensor strength was 77.0-86.5% across the post-operative timeline, only 27.6% (1 year), 46.6% (2 years) and 41.4% (5 years) of patients demonstrated an LSI ≥ 90%.

Post-operative isokinetic knee extensor strength deficits have been observed following ACI previously,[20] as well as following ACL reconstruction,[13, 16, 36] high tibial osteotomy[15] and arthroscopic partial meniscectomy.[12, 35, 37] Several factors may affect the size of post-operative isokinetic strength deficit and subsequent recovery following surgery. A longer delay between injury and surgery in ACL reconstructed patients has been associated with reduced post-operative isokinetic strength in the knee flexors and extensors.[17] Articular cartilage repair patients have been likened to osteoarthritic patients, whereby symptoms (and pain) experienced persist over a prolonged period of time.[50] Certainly, the mean duration of symptoms in this analysis was 9.3 years, unlike those following ACL reconstruction who generally experience an acute trauma and undergo immediate reconstruction. This, in combination with the need for an additional arthroscopic biopsy required for cell culturing, would contribute to the significant pre-operative knee extensor deficiencies observed during the strength assessment, and delayed post-operative recovery.

While peak knee extensor (and flexor) torque on the operated limb continued to improve at every post-operative time point, so too did isokinetic strength on the non-operated limb at the majority of post-operative intervals. In particular, knee extensor strength on the non-operated limb improved between every time point apart from 2 to 5 years (significantly between 1 and 2 years), while knee flexor strength limb improved between every time point (significantly between 2 and 5 years). This improvement in strength for the non-operated side may reflect a general increase in activity and a good post-operative outcome for the patient, though it did mean the relative change in LSI improvement for knee extensor/flexor strength over time was not as high as that for absolute knee extensor/flexor strength.

Alternatively, this may reflect a general level of pre-operative deconditioning highlighting the aforementioned issues of the lengthy duration of symptoms and the additional arthroscopic surgery required for cell harvest. In combination with the relative decrease in activity that may occur, these variables may act to promote pre-operative physical deterioration in knee function, which could also affect strength and function in the unaffected limb. While further research is required, this may suggest that pre-operative physical preparation and post-operative rehabilitation should also target the non-operated side, though still with an underlying focus on restoring limb strength symmetry.

Physical therapy and recovery time are primary factors in the improvement in the LSI following ACL reconstruction.[51] It is generally agreed that patients who do not follow structured advice and rehabilitation guidelines following MACI have an increased risk of failure, and could also demonstrate greater post-operative strength deficits. Based on the outcomes of this analysis, it would appear that the 12-week rehabilitation program and ongoing activity advice provided in this cohort was not adequate in restoring optimal knee extensor strength and limb symmetry, despite the continual improvements in knee extensor strength throughout the post-operative timeline. While varied means of developing ongoing quadriceps strength are available, due to the lengthy period required for graft maturation following MACI,[52] these patients are often limited in the intensity of quadriceps strengthening activity they can undertake even up until 12 months post-surgery. Furthermore, factors such as reduced patient compliance with time,[38] the unexpected length and demand of the rehabilitation process[53] and the unexpected additional cost required of continuing with supervised care, may be factors that contribute to the discontinuation of rehabilitation after the initial intensive 12 weeks. Interestingly, significant improvements in knee extensor strength were documented between 1 and 2 years post-surgery which, despite a lack of documentation on specific patient activity, may more likely reflect the general recovery time and gradual return to normal activities, rather than ongoing supervised intensive rehabilitation. While ongoing improvement was observed (particularly between 1 and 2 years), the persistent difference between the operated and non-operated limbs presents a demand for developing longer-term rehabilitation guidelines (1-2 year) to ensure a more optimal knee strength profile and long-term patient outcome. Alternatively, a more intensive program has been shown to return athletes to competition at a faster rate after chondrocyte implantation,[54] though the increased time and money commitments may prove difficult for many patients. Nevertheless, despite the lack of supportive evidence at present, patients need to be well educated that the recovery of optimal limb strength symmetry may take time, and returning to more demanding activity prior to this time may increase the risk of future graft and/or subsequent knee joint injury.

At all pre- and post-operative time points, there were no significant differences between the operated and non-operated limbs in the 3RM-SLR. Furthermore, the LSIs for the 3RM-SLR were ≥ 90 at all pre- and post-operative time points. Pre-operatively, this may reflect the irrelevance of this clinical strength test in patients with articular cartilage defects. Post-surgery, this may also reflect the irrelevance of this test after MACI and knee-based surgeries in general. However, this may also reflect the early introduction of hip flexor strengthening activities that can be undertaken without compromising the early repair after MACI.

While a significant difference was observed between the operated and non-operated limbs in the peak knee flexor torque pre-operatively and at 1 year post-surgery, there were no further differences following this time. Despite the significant difference in peak knee flexor torque at 1 year, the LSIs for knee flexor torque still remained ≥ 90 at all post-operative time points.

Therefore, while these patients demonstrated deficient knee flexor strength pre-surgery that continued at 1 year post-surgery, in the absence of knee pain, they had recovered full knee flexor strength by 2 years.

The H/Q ratios were significantly higher on the operated limb at all post-operative time points, compared with the non-operated limb, reflective of the significantly lower peak knee extensor torque at all time points. These ranged from 0.89-1.01 (operated limb) and 0.75-0.79 (non-operated limb), irrespective of the assessment time, compared with the 0.52 (males) and 0.61 (females) previously reported for healthy participants.[55] While expected for the operated limb, the higher pre-operative H/Q ratio observed in the non-operated limb may reflect general deconditioning of the quadriceps due to the aforementioned factors such as the relative inactivity that occurs in managing a knee with an articular cartilage defect, and a long duration of symptoms as observed in this cohort (average 9.3 years). The higher post-operative H/Q ratios in the non-operated limb may further reflect this, as well as the additional lengthy period of forced inactivity that may be required following a procedure like MACI. Again, further research is required to investigate the potential importance of concomitant physical rehabilitation for the non-operated side.

A successful return to sport is dependent on the return of muscular strength and function,[10] which has also has been suggested as a predictor of the development of knee osteoarthritis.[56] However, the clinical significance of reduced knee extensor torque values in this study remains unknown. Peak torque was not correlated with pain during the strength assessment at any of the pre- or post-operative time points, which is supported by recent research demonstrating only a weak association at best between pain and quadriceps muscle strength assessment.[57] While the purpose of this analysis was not to evaluate the association between strength and patient reported outcomes, rather whether knee strength and limb symmetry had returned to an acceptable range after MACI, previous research has shown a correlation between reduced knee extensor torque and associated self-reported scores and functional tests in patients following other knee surgeries.[35, 36, 58-60] This remains an area of future research.

A number of limitations existed in this study. Firstly, we acknowledge there are limitations to isokinetic strength testing. It assesses torque at a constant velocity, requires a reduced degree of patient control and almost exclusively evaluates concentric muscular contractions.[29] There are a number of functional tests reported;[29, 30, 61, 62] however, this study aimed to only assess isokinetic knee strength which has still been used extensively in the assessment of neuromuscular performance to identify deficits in several types of knee injury or surgery.[11-16, 20-22] Furthermore, we chose to evaluate strength in the affected limb in comparison to the contralateral limb throughout the pre- and post-operative timeline, as required for the LSI, rather than a comparison to a single measure of pre-operative strength (in the affected limb), immediate post-operative strength (in the unaffected limb) or a matched healthy cohort. However, pre-operative strength scores in the affected limb are typically not a good indication of optimal strength due to pain and muscular inhibition, as well as the lengthy duration of symptoms as demonstrated in this study. Furthermore, we observed an improvement in strength on the unaffected limb after surgery providing an increasing target for the operated side to reach. However, we do feel that using the unaffected limb immediately post-surgery as a comparative measure is biased, given it also becomes deconditioned as a result of the lengthy pre-operative duration of symptoms and the associated inactivity. Comparing the operated and non-operated limbs appears valid in assessing post-operative rehabilitation outcomes.[11-16, 20]

Secondly, we chose only to assess isokinetic peak torque. While peak torque has become the standard measure in isokinetic testing[63, 64] and has been used routinely in isokinetic strength assessment,[11, 12, 15, 16, 21, 63, 64] the torque produced at a knee angle of 45°[13] provides an alternative measure as a means of avoiding torque 'overshoot', which may provide an unrealistic representation of the true torque capabilities of the test limb. Furthermore, we only assessed strength at a single speed of 90˚/sec, despite a wide array of test speeds being reported in the literature.[11-13, 15, 16, 20, 21] We felt that a single test speed of 90˚/sec would not overly fatigue the patient as may be the case with multiple conditions, whilst providing a compromise between slow and demanding speeds around 30˚/sec that may prove difficult for MACI patients at 1 year and faster speeds around 240˚/sec which are used more commonly in endurance based assessment.[20]

Thirdly, we acknowledge there are limitations in using LSIs that have been previously reported.[61] The LSIs calculated in this study compare the operated and non-operated limbs. While all patients reported no recollection of past or recent injury and/or pathology on the contralateral limb, we cannot assume the non-operated side was 'normal', especially given the potential presence of pre-operative general deconditioning that may well arise from a mean duration of pre-operative symptoms of 9.3 years. Furthermore, while prior research on LSIs in healthy unaffected subjects is limited, limb asymmetry has been demonstrated in healthy subjects without history of lower limb injury. While Ostenberg et al. demonstrated no difference in healthy female soccer players when comparing dominant and non-dominant limbs during isokinetic knee extension strength assessment, a significant difference did exist when comparing the weakest and strongest limb.[27] Albeit, the LSIs reported were 92 and 94% for isokinetic knee extension at 60° and 180°/s, respectively, which are above the 85 and 90% thresholds employed in this study.

Fourthly, much of the current evidence for the use of isokinetic knee strength and/or functional assessment has been developed with ACL reconstruction in mind, and there is scarce research available for cartilage repair (and MACI). However, given the need for knee strength and support in the long-term health of the graft and knee joint itself, as well as minimizing the risk of subsequent injury, we would assume that these LSI guidelines would prove relevant in providing a more comprehensive evaluation of the physical condition of the patient in advocating a return to activity after MACI.

FUTURE RESEARCH DIRECTIONS

As outlined throughout the manuscript, a number of primary areas require further research to better evaluate the true relevance of reduced knee strength in patients following MACI and knee surgery in general. Firstly, given that it has been reported that an LSI below 85-90% may suggest that an individual is unsafe to return to regular sports activity,[10, 29-32] the association between LSI measures and the actual return to sport and re-injury risk should be evaluated. Secondly, as noted earlier further research is required to investigate the potential importance of concomitant physical rehabilitation for the unaffected limb both pre- and post-operatively. Finally, given the reduced LSIs in this study it would appear that the early supervised rehabilitation phase following MACI (12 weeks) is not sufficient to restore long-term knee extensor strength. Future research should evaluate the effect of more intensive structured rehabilitation from 3-12 months, as well as the effect a mid-term (1-2 year) rehabilitation intervention, on the long-term recovery of knee strength, capacity to return to sport, risk of re-injury and longer-term knee degenerative changes.

CONCLUSION

Our hypotheses were partially supported. While peak knee extensor torque was significantly lower in the operated limb at 1 year post-surgery, it was also lower at 2 and 5 years, while no differences were observed in peak knee flexor torque after 1 year post-surgery. Furthermore, while a mean LSI ≤ 90% was not observed for peak knee flexor torque at any post-operative time point, it was observed for peak knee extensor torque at all pre- and post-operative time points. A principle finding of this analysis is that patients in this study at 5 years post-surgery still presented with an LSI ≤ 90%, with almost 50% of patients demonstrating an LSI ≤ 85%. While the association between reduced knee extensor strength and inadequate limb asymmetry, and subsequent graft and/or knee re-injury is unknown, the assessment of these LSIs may allow the clinician to better evaluate the physical preparedness of the patient to return to activity based on a frequently reported criterion of assessment - the LSI for isokinetic knee strength.

REFERENCES

1. Basad E, Ishaque B, Bachmann G, Sturz H, Steinmeyer J. Matrix-induced autologous chondrocyte implantation versus microfracture in the treatment of cartilage defects of the knee: a 2-year randomised study. *Knee Surg Sports Traumatol Arthrosc.* 2010;18(4):519-527.

2. Behrens P, Bitter T, Kurz B, Russlies M. Matrix-associated autologous chondrocyte transplantation/implantation (MACT/MACI) - 5-year follow-up. *Knee.* 2006;13(3):194-202.

3. Ebert JR, Robertson WB, Woodhouse J, et al. Clinical and magnetic resonance imaging-based outcomes to 5 years after matrix-induced autologous chondrocyte implantation to address articular cartilage defects in the knee. *Am J Sports Med.* 2011;39(4):753-763.

4. Elder SH, Goldstein SA, Kimura JH, Soslowsky LJ, Spengler DM. Chondrocyte differentiation is modulated by frequency and duration of cyclic compressive loading. *Annals of Biomedical Engineering.* 2001;29(6):476-482.

5. Hunter CJ, Imler SM, Malaviya P, Nerem RM, Levenston ME. Mechanical compression alters gene expression and extracellular matrix synthesis by chondrocytes cultured in collagen I gels. *Biomaterials.* 2002;23(4):1249-1259.

6. Vunjak-Novakovic G, Martin I, Obradovic B, et al. Bioreactor cultivation conditions modulate the composition and mechanical properties of tissue-engineered cartilage. *Journal of Orthopaedic Research.* 1999;17(1):130-138.

7. Bartlett W, Skinner JA, Gooding CR, et al. Autologous chondrocyte implantation versus matrix-induced autologous chondrocyte implantation for osteochondral defects of the knee: a prospective, randomised study. *J Bone Joint Surg Br.* 2005;87(5):640-645.

8. D'Anchise R, Manta N, Prospero E, Bevilacqua C, Gigante A. Autologous implantation of chondrocytes on a solid collagen scaffold: clinical and histological outcomes after two years of follow-up. *J Orthop Traumatol.* 2005;6:36-43.

9. Ebert JR, Fallon M, Zheng MH, Wood DJ, Ackland TR. A randomized trial comparing accelerated and traditional approaches to postoperative weightbearing rehabilitation after matrix-induced autologous chondrocyte implantation: findings at 5 years. *Am J Sports Med.* 2012;40(7):1527-1537.

10. Thomee R, Kaplan Y, Kvist J, et al. Muscle strength and hop performance criteria prior to return to sports after ACL reconstruction. *Knee Surg Sports Traumatol Arthrosc.* 2011;19(11):1798-1805.

11. Campbell DE, Glenn W. Rehabilitation of knee flexor and knee extensor muscle strength in patients with meniscectomies, ligamentous repairs, and chondromalacia. *Phys Ther.* 1982;62(1):10-15.

12. Gapeyeva H, Paasuke M, Ereline J, Pintsaar A, Eller A. Isokinetic torque deficit of the knee extensor muscles after arthroscopic partial meniscectomy. *Knee Surg Sports Traumatol Arthrosc.* 2000;8(5):301-304.

13. Harter RA, Osternig LR, Singer KM, James SL, Larson RL, Jones DC. Long-term evaluation of knee stability and function following surgical reconstruction for anterior cruciate ligament insufficiency. *Am J Sports Med.* 1988;16(5):434-443.

14. Jenkins DG, Imms FJ, Prestidge SP, Small GI. Muscle strength before and after menisectomy: a comparison of methods of post-operative management. *Rheumatol Rehabil.* 1976;15(3):153-155.

15. Kawazoe T, Takahashi T. Recovery of muscle strength after high tibial osteotomy. *J Orthop Sci.* 2003;8(2):160-165.

16. Keays SL, Bullock-Saxton J, Keays AC, Newcombe P. Muscle strength and function before and after anterior cruciate ligament reconstruction using semitendonosus and gracilis. *Knee.* 2001;8(3):229-234.

17. Yuksel HY, Erkan S, Uzun M. Factors affecting isokinetic muscle strength before and after anterior cruciate ligament reconstruction. *Acta Orthop Belg.* 2011;77(3):339-348.

18. Fernandes TL, Protta TR, Fregni F, et al. Isokinetic muscle strength and knee function associated with double femoral pin fixation and fixation with interference screw in anterior cruciate ligament reconstruction. *Knee Surg Sports Traumatol Arthrosc.* 2012;20(2):275-280.

19. Schroer WC, Diesfeld PJ, Reedy ME, LeMarr AR. Isokinetic strength testing of minimally invasive total knee arthroplasty recovery. *J Arthroplasty.* 2010;25(2):274-279.

20. Loken S, Ludvigsen TC, Hoysveen T, Holm I, Engebretsen L, Reinholt FP. Autologous chondrocyte implantation to repair knee cartilage injury: ultrastructural evaluation at 2 years and long-term follow-up including muscle strength measurements. *Knee Surg Sports Traumatol Arthrosc.* 2009.

21. Van Assche D, Staes F, Van Caspel D, et al. Autologous chondrocyte implantation versus microfracture for knee cartilage injury: a prospective randomized trial, with 2-year follow-up. *Knee Surg Sports Traumatol Arthrosc.* 2010;18(4):486-495.

22. Ebert JR, Lloyd DG, Wood DJ, Ackland TR. Isokinetic knee extensor strength deficit following matrix-induced autologous chondrocyte implantation. *Clin Biomech (Bristol, Avon).* 2012;27(6):588-594.

23. Thomee R, Neeter C, Gustavsson A, et al. Variability in leg muscle power and hop performance after anterior cruciate ligament reconstruction. *Knee Surg Sports Traumatol Arthrosc.* 2012;20(6):1143-1151.

24. Barber-Westin SD, Noyes FR. Factors used to determine return to unrestricted sports activities after anterior cruciate ligament reconstruction. *Arthroscopy.* 2011;27(12):1697-1705.

25. Barber SD, Noyes FR, Mangine RE, McCloskey JW, Hartman W. Quantitative assessment of functional limitations in normal and anterior cruciate ligament-deficient knees. *Clin Orthop Relat Res.* 1990(255):204-214.

26. Petschnig R, Baron R, Albrecht M. The relationship between isokinetic quadriceps strength test and hop tests for distance and one-legged vertical jump test following anterior cruciate ligament reconstruction. *J Orthop Sports Phys Ther.* 1998;28(1):23-31.

27. Ostenberg A, Roos E, Ekdahl C, Roos H. Isokinetic knee extensor strength and functional performance in healthy female soccer players. *Scandinavian journal of medicine & science in sports.* 1998;8(5 Pt 1):257-264.

28. Augustsson J, Thomee R, Karlsson J. Ability of a new hop test to determine functional deficits after anterior cruciate ligament reconstruction. *Knee Surg Sports Traumatol Arthrosc.* 2004;12(5):350-356.

29. Juris PM, Phillips EM, Dalpe C, Edwards C, Gotlin RS, Kane DJ. A dynamic test of lower extremity function following anterior cruciate ligament reconstruction and rehabilitation. *J Orthop Sports Phys Ther.* 1997;26(4):184-191.

30. Risberg MA, Holm I, Ekeland A. Reliability of functional knee tests in normal athletes. *Scandinavian journal of medicine & science in sports.* 1995;5(1):24-28.

31. Mattacola CG, Perrin DH, Gansneder BM, Gieck JH, Saliba EN, McCue FC, 3rd. Strength, Functional Outcome, and Postural Stability After Anterior Cruciate Ligament Reconstruction. *Journal of athletic training.* 2002;37(3):262-268.

32. Noyes FR, Barber SD, Mangine RE. Abnormal lower limb symmetry determined by function hop tests after anterior cruciate ligament rupture. *Am J Sports Med.* 1991;19(5):513-518.

33. Eriksson E. Rehabilitation of muscle function after sport injury - major problem in sports medicine. *Int J Sports Med.* 1981;2(1):1-6.

34. Moffet H, Richards CL, Malouin F, Bravo G. Factors predicting knee function in patients with meniscal lesions. *J Electromyogr Kinesiol.* 1994;4(4):205-217.

35. Ericsson YB, Roos EM, Dahlberg L. Muscle strength, functional performance, and self-reported outcomes four years after arthroscopic partial meniscectomy in middle-aged patients. *Arthritis Rheum.* 2006;55(6):946-952.

36. Lautamies R, Harilainen A, Kettunen J, Sandelin J, Kujala UM. Isokinetic quadriceps and hamstring muscle strength and knee function 5 years after anterior cruciate ligament reconstruction: comparison between bone-patellar tendon-bone and hamstring tendon autografts. *Knee Surg Sports Traumatol Arthrosc.* 2008;16(11):1009-1016.

37. Sturnieks DL, Besier TF, Hamer PW, et al. Knee strength and knee adduction moments following arthroscopic partial meniscectomy. *Med Sci Sports Exerc.* 2008;40(6):991-997.

38. Beynnon BD, Johnson RJ, Fleming BC. The science of anterior cruciate ligament rehabilitation. *Clin Orthop Relat Res.* 2002(402):9-20.

39. Appell HJ. Skeletal muscle atrophy during immobilization. *Int J Sports Med.* 1986;7(1):1-5.

40. Brittberg M, Lindahl A, Nilsson A, Ohlsson C, Isaksson O, Peterson L. Treatment of deep cartilage defects in the knee with autologous chondrocyte transplantation. *N Engl J Med.* 1994;331(14):889-895.

41. Minas T, Peterson L. Autologous chondrocyte implantation. *Op Tech in Orth.* 1997;7(4):323-333.

42. Sterne JA, Davey Smith G. Sifting the evidence-what's wrong with significance tests? *Bmj.* 2001;322(7280):226-231.

43. Ageberg E, Thomee R, Neeter C, Silbernagel KG, Roos EM. Muscle strength and functional performance in patients with anterior cruciate ligament injury treated with training and surgical reconstruction or training only: a two to five-year followup. *Arthritis Rheum.* 2008;59(12):1773-1779.

44. Itoh H, Kurosaka M, Yoshiya S, Ichihashi N, Mizuno K. Evaluation of functional deficits determined by four different hop tests in patients with anterior cruciate ligament deficiency. *Knee Surg Sports Traumatol Arthrosc.* 1998;6(4):241-245.

45. Lee DY, Karim SA, Chang HC. Return to sports after anterior cruciate ligament reconstruction - a review of patients with minimum 5-year follow-up. *Annals of the Academy of Medicine, Singapore.* 2008;37(4):273-278.

46. Anderson JL, Lamb SE, Barker KL, Davies S, Dodd CA, Beard DJ. Changes in muscle torque following anterior cruciate ligament reconstruction: a comparison between hamstrings and patella tendon graft procedures on 45 patients. *Acta Orthop Scand.* 2002;73(5):546-552.

47. Arangio GA, Chen C, Kalady M, Reed JF, 3rd. Thigh muscle size and strength after anterior cruciate ligament reconstruction and rehabilitation. *J Orthop Sports Phys Ther.* 1997;26(5):238-243.

48. Carter TR, Edinger S. Isokinetic evaluation of anterior cruciate ligament reconstruction: hamstring versus patellar tendon. *Arthroscopy.* 1999;15(2):169-172.

49. Nakayama Y, Shirai Y, Narita T, Mori A, Kobayashi K. Knee functions and a return to sports activity in competitive athletes following anterior cruciate ligament reconstruction. *Journal of Nippon Medical School = Nippon Ika Daigaku zasshi.* 2000;67(3):172-176.

50. Roos E, Engelhart L, Ranstam J, et al. ICRS Recommendation Document : Patient-Reported Outcome Instruments for Use in Patients with Articular Cartilage Defects. *Cartilage.* 2011;2(2):122-136.

51. Kobayashi A, Higuchi H, Terauchi M, Kobayashi F, Kimura M, Takagishi K. Muscle performance after anterior cruciate ligament reconstruction. *Int Orthop.* 2004;28(1):48-51.

52. Edwards PK, Ackland T, Ebert JR. Clinical Rehabilitation Guidelines for Matrix-induced Autologous Chondrocyte Implantation (MACI) on the Tibiofemoral Joint. *J Orthop Sports Phys Ther.* 2013.

53. Heijne A, Axelsson K, Werner S, Biguet G. Rehabilitation and recovery after anterior cruciate ligament reconstruction: patients' experiences. *Scandinavian journal of medicine & science in sports.* 2008;18(3):325-335.

54. Della Villa S, Kon E, Filardo G, et al. Does intensive rehabilitation permit early return to sport without compromising the clinical outcome after arthroscopic autologous chondrocyte implantation in highly competitive athletes? *Am J Sports Med.* 2010;38(1):68-77.

55. Hewett TE, Myer GD, Zazulak BT. Hamstrings to quadriceps peak torque ratios diverge between sexes with increasing isokinetic angular velocity. *J Sci Med Sport.* 2008;11(5):452-459.

56. Thorstensson CA, Petersson IF, Jacobsson LT, Boegard TL, Roos EM. Reduced functional performance in the lower extremity predicted radiographic knee osteoarthritis five years later. *Ann Rheum Dis.* 2004;63(4):402-407.

57. Riddle DL, Stratford PW. Impact of pain reported during isometric quadriceps muscle strength testing in people with knee pain: data from the osteoarthritis initiative. *Phys Ther.* 2011;91(10):1478-1489.

58. Wilk KE, Romaniello WT, Soscia SM, Arrigo CA, Andrews JR. The relationship between subjective knee scores, isokinetic testing, and functional testing in the ACL-reconstructed knee. *J Orthop Sports Phys Ther.* 1994;20(2):60-73.

59. Sachs RA, Daniel DM, Stone ML, Garfein RF. Patellofemoral problems after anterior cruciate ligament reconstruction. *Am J Sports Med.* 1989;17(6):760-765.

60. Sekiya I, Muneta T, Ogiuchi T, Yagishita K, Yamamoto H. Significance of the single-legged hop test to the anterior cruciate ligament-reconstructed knee in relation to muscle strength and anterior laxity. *Am J Sports Med.* 1998;26(3):384-388.

61. Clark NC. Functional performance testing following knee ligament injury. *Physical Therapy In Sport.* 2001;2:91-105.

62. Risberg MA, Ekeland A. Assessment of functional tests after anterior cruciate ligament surgery. *J Orthop Sports Phys Ther.* 1994;19(4):212-217.

63. Kannus P. Isokinetic evaluation of muscular performance: implications for muscle testing and rehabilitation. *Int J Sports Med.* 1994;15 Suppl 1:S11-18.

64. Sapega AA. Muscle performance evaluation in orthopaedic practice. *J Bone Joint Surg Am.* 1990;72(10):1562-1574.

FIGURE LEGENDS

FIGURE 1. The progression of (A) the three-repetition maximum straight leg raise (3RM-SLR), (B) peak knee extension torque, (C) peak knee flexion torque and (D) hamstring/quadriceps (H/Q) ratio over the assessment time line. Shown are means (SE).

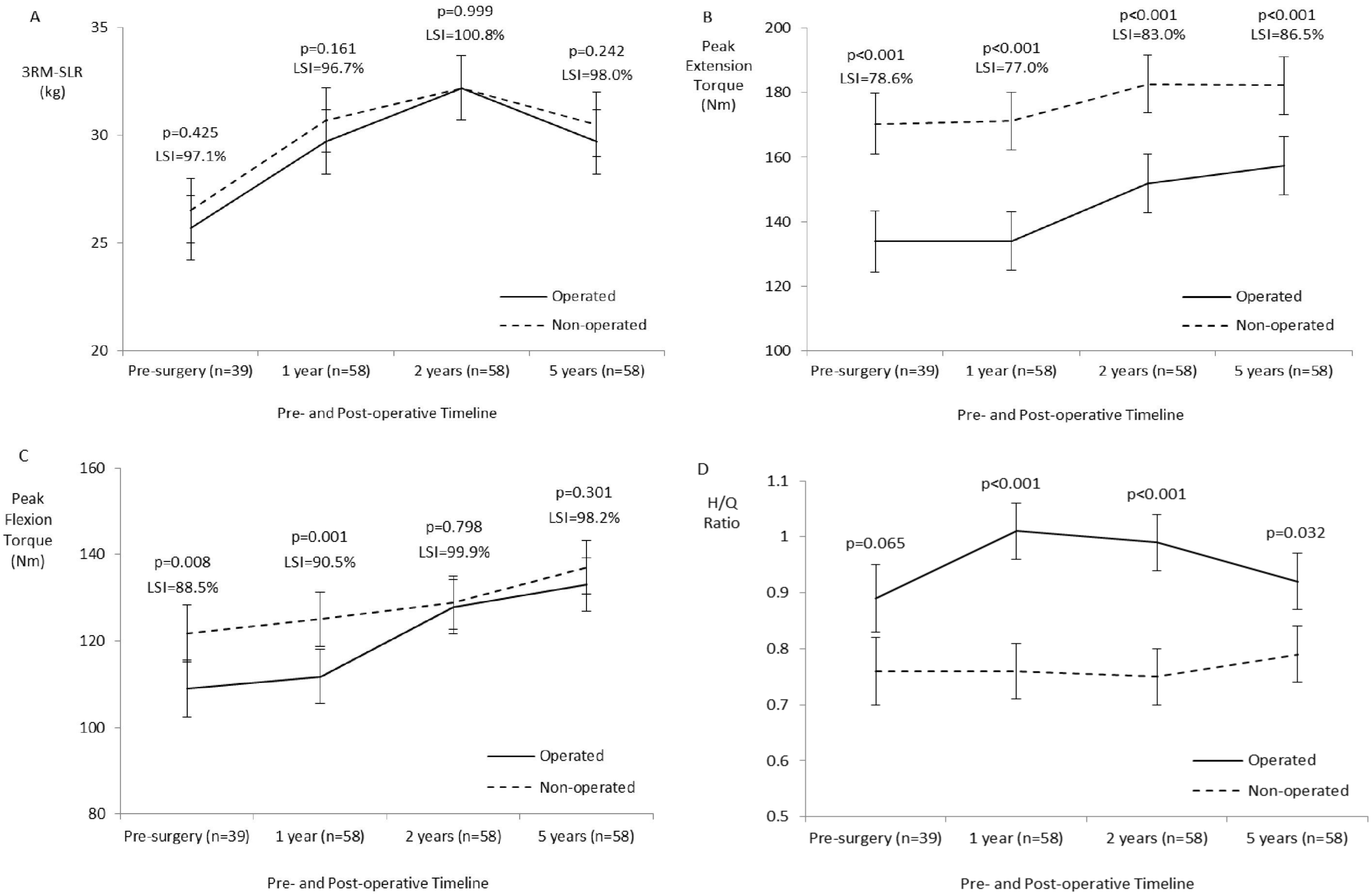
A
3RM-SLR (kg)
35
30
25
20
p=0.425
LSI=97.1%
p=0.161
LSI=96.7%
p=0.999
LSI=100.8%
p=0.242
LSI=98.0%
Operated
Non-operated
Pre-surgery (n=39)
1 year (n=58)
2 years (n=58)
5 years (n=58)
Pre- and Post-operative Timeline
B
Peak Extension Torque (Nm)
200
180
160
140
120
100
p<0.001
LSI=78.6%
p<0.001
LSI=77.0%
p<0.001
LSI=83.0%
p<0.001
LSI=86.5%
Operated
Non-operated
Pre-surgery (n=39)
1 year (n=58)
2 years (n=58)
5 years (n=58)
Pre- and Post-operative Timeline
C
Peak Flexion Torque (Nm)
160
140
120
100
80
p=0.008
LSI=88.5%
p=0.001
LSI=90.5%
p=0.798
LSI=99.9%
p=0.301
LSI=98.2%
Operated
Non-operated
Pre-surgery (n=39)
1 year (n=58)
2 years (n=58)
5 years (n=58)
Pre- and Post-operative Timeline
D
H/Q Ratio
1.1
1
0.9
0.8
0.7
0.6
0.5
p=0.065
p<0.001
p<0.001
p=0.032
Operated
Non-operated
Pre-surgery (n=39)
1 year (n=58)
2 years (n=58)
5 years (n=58)
Pre- and Post-operative Timeline

TABLE 1. The progression of post-operative weight bearing (WB), knee range of motion (ROM) and exercise rehabilitation undertaken by the MACI patient group.

Timeline	Rehabilitation Guidelines
Week 1-2	• WB Status: ≤ 20% BW • Ambulatory Aids: 2 crutches used at all times • Knee ROM: passive & active ROM from 0-30° • Knee Bracing: 0-30° • Treatment/Rehabilitation: isometric contractions & circulation exercises, CPM & cryotherapy
Week 3-6	• WB Status: 30% BW (week 3) to 60% BW (week 6) • Ambulatory Aids: 2 crutches used at all times • Knee ROM: active ROM from 0-90° (week 3) to 0-125° (week 6) • Knee Bracing: 0-45° (week 3) to full knee flexion (week 6) • Treatment/Rehabilitation: isometric/straight leg & passive/active knee flexion exercises, remedial massage, patella mobilisation, CPM, cryotherapy & hydrotherapy
Week 7-12	• WB Status: 60% BW (week 6) to full WB as tolerated (week 8) • Ambulatory Aids: 1 crutch as required until full weight bearing achieved • Knee ROM: Full active ROM (week 7) • Knee Bracing: Full knee flexion • Rehabilitation: introduction of proprioceptive/balance activities, cycling, walking, resistance & CKC activities
3-6 months	• Rehabilitation: introduction of more demanding OKC (terminal leg extension) & CKC (i.e. leg press, squats), upright cycling, rowing ergometry & elliptical trainers
6-9 months	• Rehabilitation: increase difficulty of proprioceptive/balance, OKC & CKC exercises (ie. step ups/downs, squats, lunges), introduce controlled mini trampoline jogging
9-12 months	• Rehabilitation: increase difficulty of CKC exercises (ie. lunge and squat activities on unstable surfaces), introduction of agility drills relevant to patient's sport, return to competitive activity if warranted after 12 months

ROM = range of motion; BW = body weight; WB = weight bearing; CPM = continuous passive motion; CKC = closed kinetic chain; OKC = open kinetic chain.

TABLE 2. Pearson's correlation coefficients for each of the strength measures with patient demographics (age, height, weight, body mass index), defect parameters (size), injury/surgery history (prior procedures, duration of symptoms) and pain scores (VAS) pre-operatively and at 1, 2 and 5 years post-surgery.

Strength Variable		Age	Height	Weight	Body Mass Index	Defect Size	Duration of Symptoms	Prior Procedures	Pain (VAS)
3RM-SLR	Pre-surgery (n=37)	-0.16	0.52†	0.45†	0.15	0.10	-0.13	0.16	-0.08
Peak Knee Extension Torque		-0.17	0.47†	0.37‡	0.14	-0.06	0.00	-0.46†	-0.12
Peak Knee Flexion Torque		-0.09	0.57†	0.53†	0.24	-0.05	-0.13	-0.19	-0.26
Hamstrings/Quadriceps Ratio		0.16	0.07	0.11	0.07	0.08	-0.32	0.52†	-0.20
3RM-SLR	1 year (n=58)	-0.30‡	0.55*	0.39†	0.02	0.14	-0.28‡	0.13	-0.02
Peak Knee Extension Torque		-0.29‡	0.66*	0.40†	0.06	0.05	-0.24	-0.23	0.03
Peak Knee Flexion Torque		-0.19	0.57*	0.43†	0.06	0.07	-0.26	-0.25	0.01
Hamstrings/Quadriceps Ratio		0.01	-0.37†	-0.13	0.16	-0.09	-0.06	-0.03	-0.01
3RM-SLR	2 years (n=58)	-0.23	0.57*	0.47*	0.10	0.22	-0.32‡	0.03	-0.13
Peak Knee Extension Torque		-0.38†	0.63*	0.39†	-0.05	-0.01	-0.22	0.20	0.06
Peak Knee Flexion Torque		-0.17	0.68*	0.65*	0.21	0.08	-0.32‡	-0.25	0.10
Hamstrings/Quadriceps Ratio		0.25	-0.09	0.13	0.22	0.11	-0.11	-0.12	-0.10
3RM-SLR	5 years (n=58)	-0.35‡	0.54*	0.39†	0.03	0.14	-0.27‡	0.09	0.01
Peak Knee Extension Torque		-0.32‡	0.71*	0.47†	0.00	0.09	-0.22	0.17	0.02
Peak Knee Flexion Torque		-0.08	0.72*	0.67*	0.21	0.15	-0.27‡	0.19	-0.04
Hamstrings/Quadriceps Ratio		0.30‡	-0.12	0.04	0.14	0.06	-0.09	-0.11	-0.11

$P < 0.0001$*; $P < 0.01$†; $P < 0.05$‡
3RM-SLR = three repetition maximum straight leg raise; VAS = visual analogue scale.

TABLE 3. Isokinetic knee extension (Nm) and Limb Symmetry Index: estimated within-person differences by side (operated – Op; non-operated - Nop) and time point (pre and 1, 2 and 5 years post-surgery).

Time-point	Mean (SE)		Contrast	Difference	95% CI	p-value
Isokinetic knee extension (Nm)						
	Time (Op)					
Pre (Op)	133.9*	(9.5)				
1 year (Op)	134.0	(9.0)	Pre - 1year	0.1	-10.9 - 11.0	0.988
2 years (Op)	151.9	(9.0)	1 - 2 years	17.9	8.4 - 27.4	<0.001
5 years (Op)	157.4	(9.0)	2 – 5 years	5.5	-3.4 - 15.0	0.255
	Time (Nop)					
Pre (Nop)	170.3*	(9.5)				
1 year (Nop)	171.2	(9.0)	Pre - 1year	0.9	-10.0 – 11.9	0.866
2 years (Nop)	182.7	(9.0)	1 - 2 years	11.5	2.0 – 21.0	0.018
5 years (Nop)	182.2	(9.0)	2 – 5 years	-0.5	-10.0 – 9.0	0.916
	Side					
Pre (Op)	133.9*	(9.5)				
Pre (Nop)	170.3*	(9.5)	Pre (Nop – Op)	36.4	24.4 - 48.3	<0.001
1 year (Op)	134.0	(9.0)				
1 year (Nop)	171.2	(9.0)	1 year (Nop – Op)	37.2	27.7 - 46.7	<0.001
2 years (Op)	151.9	(9.0)				
2 years (Nop)	182.7	(9.0)	2 years (Nop – Op)	30.8	21.3 - 40.3	<0.001
5 years (Op)	157.4	(9.0)				
5 years (Nop)	182.2	(9.0)	5 years (Nop – Op)	24.8	15.3 - 34.3	<0.001
Limb Symmetry Index						
	Time					
Pre	78.6	(3.0)				
1 year	77.0	(2.6)	Pre – 1 year	-1.6	-7.5 – 4.4	0.601
2 years	83.0	(2.6)	1 - 2 years	6.0	0.9 – 11.1	0.022
5 years	86.5	(2.6)	2 – 5 years	3.4	-1.7 – 8.5	0.187

*estimated marginal mean from linear regression model with generalized estimating equations.

TABLE 4. The percentage (and number) of patients with a Limb Symmetry Index (LSI) below 85% and 90%, comparing the operated to non-operated limbs, for the strength measures pre-operatively (n=37) and at 1, 2 and 5 years post-surgery (n=58).

Time-point	3RM Straight Leg Raise		Peak Knee Extension Torque		Peak Knee Flexion Torque	
	LSI ≤ 85%	LSI ≤ 90%	LSI ≤ 85%	LSI ≤ 90%	LSI ≤ 85%	LSI ≤ 90%
Pre-surgery	16.2% (n=6)	18.9% (n=7)	59.5% (n=22)	59.5% (n=22)	32.4% (n=12)	48.6% (n=18)
1 year	15.5% (n=9)	27.6% (n=16)	70.7% (n=41)	72.4% (n=42)	37.9% (n=22)	41.4% (n=24)
2 years	12.1% (n=7)	17.2% (n=10)	44.8% (n=26)	53.4% (n=31)	17.2% (n=10)	31.0% (n=18)
5 years	8.6% (n=5)	17.2% (n=10)	48.3% (n=28)	58.6% (n=34)	15.5% (n=9)	32.8% (n=19)

TABLE 5. Isokinetic knee flexion (Nm) and Limb Symmetry Index: estimated within-person differences by side (operated – Op; non-operated - Nop) and time point (pre and 1, 2 and 5 years post-surgery).

Time-point	Mean (SE)		Contrast	Difference	95% CI	p-value
Isokinetic knee flexion (Nm)						
Time (Op)						
Pre (Op)	109.0*	(6.6)				
1 year (Op)	111.8	(6.2)	Pre - 1year	2.8	-5.9 – 11.5	0.527
2 years (Op)	127.9	(6.2)	1 - 2 years	16.0	8.5 – 23.5	<0.001
5 years (Op)	133.0	(6.2)	2 – 5 years	5.2	-2.3 – 12.7	0.177
Time (Nop)						
Pre (Nop)	121.8*	(6.6)				
1 year (Nop)	125.0	(6.2)	Pre - 1year	3.1	-5.5 – 11.8	0.479
2 years (Nop)	128.8	(6.2)	1 - 2 years	3.9	-3.7 – 11.4	0.314
5 years (Nop)	137.0	(6.2)	2 – 5 years	8.2	0.6 – 15.7	0.033
Side						
Pre (Op)	109.0*	(6.6)				
Pre (Nop)	121.8*	(6.6)	Pre (Nop – Op)	12.8	3.4 – 22.2	0.008
1 year (Op)	111.8	(6.2)				
1 year (Nop)	125.0	(6.2)	1 year (Nop – Op)	13.1	5.6 – 20.7	0.001
2 years (Op)	127.9	(6.2)				
2 years (Nop)	128.8	(6.2)	2 years (Nop – Op)	1.0	-6.5 – 8.5	0.798
5 years (Op)	133.0	(6.2)				
5 years (Nop)	137.0	(6.2)	5 years (Nop – Op)	4.0	-6.5 – 11.5	0.301
Limb Symmetry Index						
Time						
Pre	88.5*	(3.1)				
1 year	90.5	(2.5)	Pre - 1year	2.0	-5.4 – 9.4	0.597
2 years	99.9	(2.5)	1 - 2 years	9.4	2.9 – 15.9	0.004
5 years	98.2	(2.5)	2 – 5 years	-1.6	-8.1 – 4.8	0.617

*estimated marginal mean from linear regression model with generalized estimating equations.

TABLE 6. Three-repetition maximum straight leg raise (3RM-SLR - kg) and Limb Symmetry Index: estimated within-person differences by side (operated – Op; non-operated - Nop) and time point (pre and 1, 2 and 5 years post-surgery).

Time-point	Mean (SE)		Contrast	Difference	95% CI	p-value
3RM-SLR (Kg)						
Time (Op)						
Pre (Op)	25.7*	(1.5)				
1 year (Op)	29.7	(1.5)	Pre - 1year	3.9	2.3 – 5.6	<0.001
2 years (Op)	32.2	(1.5)	1 - 2 years	2.5	1.0 – 3.9	0.001
5 years (Op)	29.7	(1.5)	2 – 5 years	-2.5	-3.9 - -1.1	0.001
Time (Nop)						
Pre (Nop)	26.5*	(1.5)				
1 year (Nop)	30.7	(1.5)	Pre - 1year	4.3	2.6 – 5.9	<0.001
2 years (Nop)	32.2	(1.5)	1 - 2 years	1.4	0.0 – 2.9	0.052
5 years (Nop)	30.5	(1.5)	2 – 5 years	-1.6	-3.1 - -0.2	0.026
Side						
Pre (Op)	25.7*	(1.5)				
Pre (Nop)	26.5*	(1.5)	Pre (Nop – Op)	0.7	-1.0 – 2.5	0.425
1 year (Op)	29.7	(1.5)				
1 year (Nop)	30.7	(1.5)	1 year (Nop – Op)	1.0	-0.4 – 2.5	0.161
2 years (Op)	32.2	(1.5)				
2 years (Nop)	32.2	(1.5)	2 years (Nop – Op)	0.0	-1.4 – 1.4	0.999
5 years (Op)	29.7	(1.5)				
5 years (Nop)	30.5	(1.5)	5 years (Nop – Op)	0.9	-0.6 – 2.3	0.242
Limb Symmetry Index						
Time						
Pre	97.1*	(2.8)				
1 year	96.7	(2.3)	Pre - 1year	-6.1	-12.7 – 0.6	0.076
2 years	100.8	(2.3)	1 - 2 years	4.2	-1.7 – 10.0	0.168
5 years	98.0	(2.3)	2 – 5 years	-2.8	-8.7 – 3.1	0.348

*estimated marginal mean from linear regression model with generalized estimating equations.

TABLE 7. Hamstring/quadriceps (H/Q) ratio: estimated within-person differences by side (operated – Op; non-operated - Nop) and time point (pre and 1, 2 and 5 years post-surgery).

Time-point	Mean (SE)		Contrast	Difference	95% CI	p-value
			Time (Op)			
Pre (Op)	0.89*	(0.06)				
1 year (Op)	1.01	(0.05)	Pre - 1year	0.11	-0.01 – 0.25	0.076
2 years (Op)	0.99	(0.05)	1 - 2 years	-0.02	-0.13 – 0.09	0.709
5 years (Op)	0.92	(0.05)	2 – 5 years	-0.07	-0.18 – 0.04	0.227
			Time (Nop)			
Pre (Nop)	0.76*	(0.06)				
1 year (Nop)	0.76	(0.05)	Pre - 1year	0.00	-0.12 – 0.13	0.941
2 years (Nop)	0.75	(0.05)	1 - 2 years	-0.01	-0.12 – 0.10	0.835
5 years (Nop)	0.79	(0.05)	2 – 5 years	0.04	-0.06 – 0.15	0.456
			Side			
Pre (Op)	0.89*	(0.06)				
Pre (Nop)	0.76*	(0.06)	Pre (Nop – Op)	-0.13	-0.27 – 0.01	0.065
1 year (Op)	1.01	(0.05)				
1 year (Nop)	0.76	(0.05)	1 year (Nop – Op)	-0.24	-0.36 - -0.13	<0.001
2 years (Op)	0.99	(0.05)				
2 years (Nop)	0.75	(0.05)	2 years (Nop – Op)	-0.23	-0.25 - -0.12	<0.001
5 years (Op)	0.92	(0.05)				
5 years (Nop)	0.79	(0.05)	5 years (Nop – Op)	-0.12	-0.23 - -0.01	0.032

*estimated marginal mean from linear regression model with generalized estimating equations.

TABLE 8. Sensitivity analysis: Differences in strength measures analyzed on an available case basis, between the operated (Op) and non-operated (Nop) limbs.

Measure	Contrast	Difference	95% CI	p-value
3RM SLR (kg)	Pre: Op vs Nop	-0.7	-2.4 – 1.0	0.406
	Op: Pre – 1 year	4.1	2.2 – 6.0	<0.001
Isokinetic Knee Extension (Nm)	Pre: Op vs Nop	36.4	21.5 - 51.2	<0.001
	Op: Pre – 1 year	1.7	-8.5 – 11.9	0.740
Isokinetic Knee Flexion (Nm)	Pre: Op vs Nop	12.8	3.8 – 21.9	0.007
	Op: Pre – 1 year	5.6	-4.0 – 15.3	0.245
H/Q Ratio	Pre: Op vs Nop	-0.13	-0.21-0.05	0.002
	Op: Pre – 1 year	0.10	-0.04 - 0.24	0.167

3RM-SLR = three-repetition maximum straight leg raise; Pre = pre-surgery; Op = operated; Nop = non-operated.