#### KNEE

# **Tibiofemoral loss of contact area but no changes in peak pressures after meniscectomy in a Lapine in vivo quadriceps force transfer model**

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#### **Abstract**

*Purpose* The menisci are thought to modulate load transfer and to absorb shocks in the knee joint. No study has experimentally measured the meniscal functions in the intact, in vivo joint loaded by physiologically relevant muscular contractions.

*Methods* Right knee joints of seven New Zealand white rabbits were loaded using isometric contractions of the quadriceps femoris muscles controlled by femoral nerve stimulation. Isometric knee extensor torques at the maximal and two submaximal force levels were performed at knee angles of 70°, 90°, 110°, and 130°. Patellofemoral and tibiofemoral contact areas and pressure distributions were measured using Fuji Presensor film inserted above and below the menisci and also with the menisci removed.

*Results* Meniscectomy was associated with a decrease in tibiofemoral contact area ranging from 30 to 70 % and a corresponding increase in average contact pressures. Contact areas measured below the menisci were consistently larger than those measured on top of the menisci. Contact areas in the patellofemoral joint (PFJ), and peak pressures in tibiofemoral and PFJs, were not affected by meniscectomy. Contact areas and peak pressures in all joints depended crucially on knee joint angle and quadriceps

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force: The more flexed the knee joint was, the larger were the contact areas and the higher were the peak pressures. *Conclusions* In agreement with the literature, removal of the menisci was associated with significant decreases in tibiofemoral contact area and corresponding increases in average contact pressures, but surprisingly, peak pressures remained unaffected, indicating that the function of the menisci is to distribute loads across a greater contact area.

**Keywords** Rabbit knee joint model · Load transfer · Contact pressure measurement · Dynamic muscle stimulation · Meniscus · Meniscectomy

#### **Introduction**

Menisci in the knee are fibro-cartilaginous structures thought to transfer loads across the tibiofemoral joints and act as shock absorbers in the knee  $[1, 33]$  $[1, 33]$  $[1, 33]$  $[1, 33]$  $[1, 33]$ . They are also implicated in joint proprioception, lubrication, and nutrition of the adjacent cartilages [\[1](#page-7-0)]. Partial meniscectomy and complete meniscectomy have been associated with an increased risk of knee osteoarthritis [\[24](#page-8-1), [30](#page-8-2), [31\]](#page-8-3), and meniscectomy in the rabbit knee is a frequently used experimental model for post-traumatic osteoarthritis (OA) [[10,](#page-7-1) [28](#page-8-4), [30](#page-8-2), [31\]](#page-8-3) that is thought to mimic the human condition well [\[11](#page-7-2), [21](#page-7-3)].

Experimental and theoretical works in cadavers suggest that the loss of the menisci results in a 30–80 % decrease in tibiofemoral contact area and a 170–400 % increase in average and peak contact pressures [\[7](#page-7-4), [34\]](#page-8-5), with the effects depending on changes of axial force during gait cycle with two peak loads at 14 and 45 % of gait cycle [[4\]](#page-7-5). Therefore, these factors were identified as risk factors developing cartilage degeneration and osteoarthritis.

Most experimental studies aimed at investigating the effects of the menisci on pressure distributions in the knee are based on research using ex vivo approaches or cadaver specimens [[4,](#page-7-5) [7,](#page-7-4) [13](#page-7-6), [34](#page-8-5)]. Furthermore, the loading conditions are simulated using robots or material testing devices and the joints are largely dissected. Although these models have provided great insight into knee joint kinematics and kinetics for boundary conditions consistent with the experimental setups, they do not represent physiological loading conditions of the intact joint loaded by muscular contractions [[4,](#page-7-5) [7](#page-7-4), [13,](#page-7-6) [34\]](#page-8-5). In order to approach physiological loading conditions, controlled muscular contractions were applied by stimulating the quadriceps muscle through a femoral nerve electrode.

Therefore, the aim of this study was to quantify tibiofemoral and patellofemoral contact areas and pressures while the knee was loaded by controlled muscular contractions prior to and following bilateral meniscectomy. Based on previously published studies and clinical experience, it was hypothesized that following meniscectomy peak pressures increase and contact areas decrease in the tibiofemoral joints, while the patellofemoral joint (PFJ) remained unaffected.

#### **Materials and methods**

In order to measure contact area and peak pressure, pressure-sensitive film was inserted into the tibiofemoral and PFJs of New Zealand white rabbits, above and below the menisci, and prior to and following meniscectomy. Measurements were made during submaximal and maximal isometric knee extensor contractions at different knee joint angles in order to simulate physiologically occurring conditions.

## Animals

Seven 1-year-old female New Zealand white rabbits (average mass  $= 5.6$  kg; range 4.7–6.5 kg, Riemens, St. Agatha, ON, Canada) were used for this experiment. Rabbits were tranquilized with 0.18 ml Atravet (25 mg/ml; Vetoquionol NA. Inc., Lavaltrie, QC, Canada) and held under anesthesia with a 2 % isoflurane/oxygen mixture. After the experiment, animals were killed with an overdose injection of Euthanyl (MTC Pharmaceuticals; Cambridge, ON, Canada) into the lateral ear vein.

#### Surgery

A custom-designed, cuff-type electrode was implanted on the femoral nerve of the right leg to allow for electrical stimulation of the knee extensor muscles [\[27](#page-8-6)]. Then, the knee joint was exposed while carefully preserving the extensor mechanism (quadriceps muscles and patellar tendon) and all ligaments so as to preserve the kinematics of normal knee joint function. The medial and lateral menisci were kept fully intact, and all meniscal insertions were preserved. The joint capsule was minimally opened to allow for gentle insertion of the pressure-sensitive film in order to not produce any artifacts [[3,](#page-7-7) [18,](#page-7-8) [34\]](#page-8-5) (Fig. [1](#page-1-0)).

Muscle stimulation and force measurements

Animals were placed in a stereotactic frame and the pelvis and femoral condyles were fixed with bone pins [\[27](#page-8-6)]. The knee center of rotation was carefully aligned with the rotational axis of a servomotor (Parker Hannifin Corporation, Irwin, PA, USA) which controlled (Motion Planner, Rohnert Park, CA, USA) the angle of the tibia and femur [\[5](#page-7-9)]. Knee joint moments were measured using a custombuilt force sensor (Vishay 2100 amplifier; Vishay Precision Group, Wendell, NC, USA) using Windaq data collection software (Dataq Instruments, Akron; collection card, DI-400, 12 bit) and a customized MATLAB program (The MathWorks, Natick, MA, USA).

Stimulation of the femoral nerve was given through a dual output stimulator (Grass S8800, Astro/Med Inc., Longueil, QC, Canada), which was synchronized with the servomotor. Rest periods of at least 1 min were given between contractions. Fatigue throughout the protocol was assessed by repeating the first torque measurement at the end of all testing.

The stimulation current was set at twice the level that was found to produce maximal forces to ensure recruitment of all motor units of the quadriceps muscle group (100 Hz)



<span id="page-1-0"></span>**Fig. 1** Experimental Setup. Fuji Film was inserted below (shown here) and above the meniscus. With the same contractions, also patellofemoral compartments were measured (not shown here)

as performed in earlier studies [[5,](#page-7-9) [6,](#page-7-10) [14,](#page-7-11) [16,](#page-7-12) [27](#page-8-6), [29](#page-8-7)]. For the two submaximal levels of activation, stimulation frequencies were adjusted by repetitive stimulations individually to produce forces of approximately 67 and 33 % of the "maximal" force, respectively.

#### Pressure measurement

Knee extensor forces and impulses, as well as tibiofemoral and patellofemoral contact areas and pressures were measured for isometric contractions at knee angles of 70°, 90°, 110°, and 130° which cover the range of motion of the knee during rabbit locomotion. More extended knee angles cannot be tested as the knee extensor muscles in rabbits become actively insufficient at about 150° (personal observations) and thus cannot be used to load the knee. Our measurement range is consistent with previous studies on the rabbit knee joint [[14,](#page-7-11) [16,](#page-7-12) [27\]](#page-8-6). According to biomechanical rules, a rectangular knee joint was defined as 90° flexed, while a fully extended knee joint was defined as 180° flexed. That means, the smaller the knee joint flexion angle, the more flexed the knee joint was.

Low- and medium-grade pressure-sensitive films (Fuji prescale film, Fuji Photo Film Co. Ltd., Tokyo, Japan) were used to assess contact areas  $\text{(mm)}^2$  and peak pressures (MPa). Strips of films were individually shaped to fit the joint size and geometry as good as possible and were sealed with polyethylene film for moisture protection [[15\]](#page-7-13). The average thickness of the sealed film packages was 0.26 mm. Separate film strips were placed into the medial tibiofemoral joint (MTFJ), lateral tibiofemoral joint (LTFJ), and PFJ. Measurements were performed with the Fuji film above the menisci (menisco-femoral compartment), below the menisci (menisco-tibial compartment), and after removal of the menisci. Following exposure, Fuji films were scanned (600 dpi) and analyzed using a customwritten MATLAB program (The MathWorks, Natick, MA, USA) [\[27](#page-8-6)]. Low-range pressure-sensitive film (pressure range of approximately 1–10 MPa) was used to measure total joint contact areas and peak pressures. If the lowrange sensitive film was saturated, trials were repeated with the medium-range sensitive film (pressure range of approximately 10–60 MPa) for peak pressure determination [\[32](#page-8-8)]. Mean pressure was calculated as the total force transferred across a joint compartment (obtained by integrating pressure over the entire contact area) divided by the total contact area.

Fuji films were calibrated by applying a set of known pressures with a flat ended indenter of 2 mm diameter attached to a materials testing machine (MTS) at intervals of 0.5 MPa covering the entire range of experimentally observed contact pressures [[29\]](#page-8-7). The resulting stain intensities were approximated using a third-order polynomial

function [\[29](#page-8-7)]. In order to quantify the possible pressure artifacts produced by film insertion or passive film motion, films were analyzed following insertion into the three compartments of the passive knee. These passive experiments never produced any measurable pressure stains. Also, every staining was checked for quality, in terms of insufficient quality (e.g. artifacts, touching of the boarder of the Film), measurements were repeated.

All procedures were approved by the IRB and Animal Care Committee of the University of Calgary (Calgary, Alberta, Canada), ID# AC11-0035.

#### Statistical analysis

Based on data from a pilot study, we were able to show a mean difference in our main endpoints (contact area and medial compartment) of  $3.8 \pm 1.75$  or  $6.9 \pm 1.9$ , consistent with a minimum effect size of 2.2. Thus, we calculated a sample size with the most conservative estimate of an effect size of 2 for a power of 85 % and an alpha of 5 % for a repeated measure *F* test (ANOVA) using intercooled Stata 10 (StataCorp LP, College Station, TX). The required total sample size for these parameters is 6, which we increased by 1 to  $n = 7$  to account for potential attrition, i.e. loss of an animal before analysis. With the maximum effect size seen in the pilot study (3.6), this sample size would give a power of 99 %. Further statistical analysis was performed using Excel 2007 (Microsoft Co., Washington, USA) and SPSS 19 (PASW Statistics, SPSS Inc., Chicago, Illinois) using one-way ANOVA. Contact area and peak pressure were taken as the primary dependent variables in our study, while pressure sensor location (above, below, and without meniscus) was treated as independent variables. Specific subgroup analyses were performed for the three compartments of the knee (medial, and lateral tibiofemoral, and patellofemoral) and for the knee joint flexion angles (70°, 90°, 110°, and 130°). For post hoc testing, the HSD Tukey test was used. The level of significance was set at  $\alpha = 0.05$ .

#### **Results**

# Torque

Knee extensor torque production was consistent throughout the experiments with no measurable fatigue. The highest knee extensor torques were obtained at knee angle of 70°, and they decreased steadily with increasing knee extension for all levels of stimulation (Fig. [2\)](#page-3-0). The highest torques were obtained for maximal stimulation and at 70° of knee joint flexion (mean  $~4.8$  Nm), while the lowest torques were noticed for the submaximal two stimulation conditions at  $130^{\circ}$  (mean ~0.3 Nm).

#### Contact area

Contact areas in the medial and lateral tibiofemoral compartments were significantly reduced following meniscectomy



<span id="page-3-0"></span>**Fig. 2** Torque Production. Torque production of the quadriceps femoris muscle induced by electrical femoral nerve stimulation at three different levels of stimulation [(a) 100 Hz (maximal stimulation); (b) 55–60 Hz (submaximal 1 stimulation); (c) 35–40 Hz (submaximal 2 stimulation)]. No significant differences (*α* < 0.05) were found within the groups. *Circle* without meniscus; *cross* above meniscus; *square* below meniscus

for all trials at corresponding knee angles and muscle forces. These findings were consistent across all stimulation levels. Contact areas below the menisci were larger than those above the menisci for all but the most flexed knee angle for the maximal activation conditions (Table [1](#page-3-1)). Post hoc testing showed significant differences at 90° for the "below meniscus" group in the medial tibiofemoral compartment compared to the "above meniscus" and "without meniscus" group. Results for submaximal 1 and submaximal 2 contractions were not conceptually different (Tables [2,](#page-4-0) [3](#page-5-0)).

Contact area was found to decrease with increasing knee extension (Table [1](#page-3-1)). Also contact areas decreased for a given joint angle with decreasing knee extensor forces. Contact areas in the patellofemoral compartment became smaller with decreasing muscular forces and increasing knee extension angles, but were not affected by meniscectomy (results not shown).

## Peak pressure

Peak pressures in all knee joint compartments (medial and lateral tibiofemoral, and PFJs) were essentially unaffected by meniscectomy and were similar whether measured above or below the menisci. Peak pressures decreased with decreasing levels of stimulation and increasing knee extension, as expected (Tables [1](#page-3-1), [2](#page-4-0), [3](#page-5-0)).



#### <span id="page-3-1"></span>**Table 1** Results of maximal stimulation testings (100 Hz)

Results for the medial and lateral tibiofemoral compartment. Results for the patellofemoral compartment are not shown (no differences between groups)

Post hoc testing noted significance between "above and without meniscus" (AW), between "below and without meniscus" (BW), and between "above and below meniscus" (AB); *n.s.* no significance in post hoc testing was found

**Table 2** Results of submax 1 testing (55–60 Hz)

Results for the medial and lateral tibiofemoral

between groups) Post hoc testing noted

post hoc testing was found

<span id="page-4-0"></span>

130 13.57 0.08 16.50 0.65 17.06 2.56 n.s. n.s.

Force transmission across knee compartments

The relative force transfer across the PFJ increased with increasing knee flexion. At a knee angle of 70°, about 45 % of the total force transmitted across the knee went through the PFJ (and only 55 % through the two tibiofemoral joints combined), while this contribution decreased to about 27 % (increased to 73 % for the tibiofemoral joints) for the most extended knee angle measured here (130°; Fig. [3\)](#page-5-1). Force transmission across the medial and LTFJs was approximately equal for all force levels and knee angles and varied around  $50 \pm 4$  %.

## **Discussion**

The most important findings of the present study were that contact areas and peak pressures in each compartment depended greatly on joint loading, quantified by the muscular knee extensor torque. Meniscectomy had a significant effect on tibiofemoral but not on patellofemoral contact areas, but did not—in contrast to our hypothesis—affect peak pressures in any of the knee compartments. Since force transmission across the three knee compartments were essentially unaffected by meniscectomy, the decreased contact areas following meniscectomy resulted in an increase in the average contact pressures that was proportional to the decrease in contact areas (results not shown).

Force transfers across the knee, and the specific role of the menisci in such transfers, has been studied extensively [\[3](#page-7-7), [7,](#page-7-4) [26](#page-8-9), [34](#page-8-5)]. To our knowledge, these experiments have been performed exclusively in human or animal cadavers where loads were applied using wires and pulleys, and lines of action, force magnitudes, and force sharing among muscles had to be estimated. In contrast, in our experiment, torques were produced by controlled electrical stimulation of the quadriceps muscles. Therefore, the joint loading occurred in a near physiological manner, force magnitudes were realistic, and lines of action of the individual quadriceps muscles were given and did not have to be estimated. The forces applied for the different experimental conditions ranged from approximately 20 N (knee joint flexion angle 130°; stimulation, submaximal 2) to 550 N (knee joint flexion angle 70°; stimulation, maximal), thereby essentially covering the entire range of possible force production by the rabbit knee extensors. As the quadriceps muscle is the only knee extensor muscle [[8\]](#page-7-14), it

<span id="page-5-0"></span>



Results for the medial and lateral tibiofemoral compartment. Results for the patellofemoral compartment are not shown (no differences between groups)

Post hoc testing noted significance between "above and without meniscus" (AW), between "below and without meniscus" (BW), and between "above and below meniscus" (AB); *n.s.* no significance in post hoc testing was found



<span id="page-5-1"></span>**Fig. 3** Force sharing among compartments for maximal quadriceps muscle stimulation. Amount of force (in %) transmitted through each knee joint compartment as a function of knee joint angle (with 95 % confidence intervals shown)

seemed feasible to use isolated quadriceps muscle stimulation in this isometric model [\[17](#page-7-15)]. For a dynamic model, it is much more difficult to mimic the complex interplay of agonistic and antagonistic muscles. Pressure measurements were performed with Fuji Prescale Pressure-sensitive film,

which has the advantage that it is thinner and more flexible than other pressure sensors, and thus was perfect for the small and curved rabbit knee. Tekscan measurements (which are available in our laboratory) were not used despite the advantage of time-resolved pressure results [[26,](#page-8-9) [34](#page-8-5)], because of its thickness (once moisture proofed) and the artifacts introduced by bending the Tekscan material in the curved compartments of the rabbit knee. Losing timeresolved pressure measurements was considered a minor drawback in this purely isometric assessment of knee joint pressure distributions.

Meniscectomy led to a 30–70 % decrease in contact area in the medial and lateral tibiofemoral compartment at all knee angles and muscle contraction levels, which agrees with findings reported in cadaver studies (30 and 80 %; [[7,](#page-7-4) [34](#page-8-5)]). Contact areas in the PFJ were not affected by meniscectomy but decreased with decreasing muscular loading, which is consistent with findings by Clark et al. [\[6](#page-7-10)].

Contact areas below the menisci were approximately 10–25 % greater than the corresponding contact areas measured above the menisci for maximal contractions of the knee extensors, indicating that the average pressure is decreased by the same amount on the tibial plateau compared to the femoral condyle. However, for submaximal levels of knee extension, this difference was not nearly as apparent, and since rabbit hopping is associated with forces

<span id="page-6-0"></span>**Fig. 4** Visual inspection of raw data stains. This figure showed examples from low pressure stains at 90° knee joint flexion of the medial tibiofemoral compartment above, below, and without meniscus. Note the much larger area of complete film saturation without the meniscus



of about 10 % of maximal [[9\]](#page-7-16), it is doubtful whether this difference in contact area and pressures above and below the menisci plays a significant role in the rabbit's everyday life.

Interestingly, peak pressures in the tibiofemoral compartments of the rabbit knee were not affected by meniscectomy for any knee angle or level of muscle contraction, while it has been assumed (based on theoretical works and cadaver studies) that a primary function of the menisci is to reduce peak contact pressures [\[7](#page-7-4)]. Increases in peak pressures of 117–400 %, depending on the specific conditions, have been reported following total meniscectomy [\[34](#page-8-5)] or anterior or posterior root detachment  $[2, 23]$  $[2, 23]$  $[2, 23]$  $[2, 23]$  in the human knee joint. These findings are based on cadaver and ex vivo testing. The basic interpretation was that the smaller area is compensated by a higher peak pressure [[19,](#page-7-19) [20\]](#page-7-20).

Since the contact area is reduced and the average pressure is increased with meniscectomy, but peak pressures remain unchanged, our results imply that in the meniscectomized joint, areas of high pressure are greater than in the intact joint. This observation is supported by inspection of our raw data pressure stains (Fig. [4\)](#page-6-0). It appears, therefore, that meniscectomy decreases tibiofemoral contact areas and increases the areas of "high" contact pressures, but does not affect the maximal value of peak pressures in the tibiofemoral compartments of the knee. Since meniscectomy is an acknowledged risk factor for the onset of OA, maybe the primary function of the menisci is to ensure a smooth gradient of pressure distributions and minimizing the area of high contact pressures (Fig. [5](#page-6-1)). Also, the onset of OA may not necessarily be associated with the magnitude of peak pressure, but the time integral of pressure at a given joint location. Following meniscectomy, when the area of high pressures is increased, a given point on the articular contact surface is likely to see higher pressures over a longer period of time, thereby potentially creating conditions for the onset of OA. Our results do not question the increased risk of osteoarthritis after meniscectomy, nor do we provide direct results supporting or rejecting a mechanical cause for increased risk following meniscectomy.



<span id="page-6-1"></span>**Fig. 5** Theoretical model of load transfer: Schematic drawing of the difference in load transfer (contact pressure) as a function of contact area for conditions with the menisci intact (*full line*) or resected (*dashed line*). The conditions shown are for equal force transfer across the joint (identical area under the pressure–area curve). *CMP* center of maximal pressure

The results of this study need careful interpretation keeping in mind its limitations. Loading of the joint was applied through isometric contractions of the knee extensors, while we would expect normal knee joint loading to occur dynamically and with a combination of isometric, concentric, and eccentric contractions. Pressures and contact areas were measured using Fuji pressure-sensitive film which has an estimated accuracy for the rabbit knee joint of about 10 % [\[35](#page-8-10)]. Therefore, only differences greater than about 10 % and occurring systematically across joint angles and force levels should be considered with confidence. Translation of our findings to the human knee also has to be made with caution. Although the knee joints of rabbits and humans are similar [\[25](#page-8-11)], subtle differences may potentially result in vastly differing results. However, the fact that we confirmed observations in the human knee of substantial and systematic decreases in tibiofemoral contact areas across all conditions following meniscectomy, suggests that

some of our results do translate to the human knee. These controversial findings are supported by new cadaveric results of meniscal reconstruction. Meniscus suture [\[22](#page-7-21)], meniscal root refixation and meniscus transplantation [[12\]](#page-7-22) cannot fully reconstruct contact area, while peak pressure is normalized. Also Bedi et al. [[4\]](#page-7-5) showed that more than 90° of meniscus needs to be resected to change peak pressures.

The fact that we did not observe the expected increases in peak pressures in the rabbit knee with meniscectomy suggests either that the human and rabbit knee differ conceptually in this aspect or that applying loads with wires and pulleys, as compared to contractions of the fully intact muscles, might be responsible for the observed differences of our results with those typically cited in the literature. For lack of evidence, we would like to suggest that it is rather the latter than the former that caused the conceptual differences in peak pressures following meniscectomy, primarily because the rabbit and human knee are much closer in geometry and function than wires and pulleys are to the actual loading produced by the intricate and complex interplay of the intact knee extensor muscles.

#### **Conclusion**

In conclusion, meniscectomy decreases contact areas, increases average contact pressures, but does not affect peak pressures in the tibiofemoral compartments of the rabbit knee. We suggest that these results also hold for human meniscectomized knees and that the increased peak pressures observed in human cadaveric studies are caused by the artificial, rather than muscular, application of joint loading. Based on that, one might conclude that the pathomechanism of cartilage degeneration and development of osteoarthritis after total meniscectomy is primarily driven by changes in contact area and average pressure distribution but is not affected by maximal peak pressure as this does not change.

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**Conflict of interest** None of the authors have anything to disclose.

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