MEDIAL MENISCUS REPLACEMENT BY A TENDON AUTOGRAFT

EXPERIMENTS IN SHEEP

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In 20 skeletally mature female merino sheep, divided into four groups, we performed total medial meniscectomy, removal of the middle third of the patellar tendon, and tenotomy of the calcaneal tendon of the right hind leg. Group I (control) had no additional procedures. In the other three groups the medial meniscus was replaced by the middle third of the patellar tendon from the ipsilateral knee. The animals were killed at three (group II), six (group III), or 12 months (group IV) and the tendon-meniscus examined macroscopically, by light and scanning electron microscopy, and biomechanically. Remodelling of the tissue had taken place by 12 months but the failure stress and tensile modulus for the tendon-meniscus were lower than for the normal meniscus.

Our evidence suggests that, in sheep, replacement of a meniscus by a tendon autograft may decrease the severity of the degenerative changes that occur after meniscectomy.

Total and subtotal meniscectomy leads to poor clinical results in the long term in many patients (Tapper and Hoover 1969; Jørgensen et al 1987) with radiological evidence of increasing degenerative joint disease (Fairbank 1948). Tibiofemoral force transmission is altered with increased stress concentration in the articular cartilage (Kettelkamp and Jacobs 1972; Seedhom, Dowson and Wright 1974; Krause et al 1976; Shrive, O'Connor and Goodfellow 1978; Kurosawa, Fukubayashi and Nakajima 1980). In experimental animals,

degenerative changes in the hyaline cartilage occur soon after meniscectomy (Lutfi 1975; Lanzer and Komenda 1990) despite regeneration of a meniscus.

If neither preservation of the peripheral part of the meniscus (Gillquist and Oretorp 1982) nor reconstruction of the damaged meniscus (Henning et al 1990) is possible, meniscal replacement needs to be considered (Keene, Paterson and Teague 1987; Milachowski et al 1988). The use of fresh (Milachowski et al 1988) or cryopreserved allografts (Arnoczky and Milachowski 1990; Arnoczky, Warren and McDevitt 1990; Tachibana 1990) has had promising results in animal experiments. Clinically, good function and survival of fresh meniscal allografts have been reported (Zukor et al 1990), but there is the risk of disease transmission. Sterilised allografts have not proved successful in animal experiments (Kohn, Milachowski and Wirth 1988). An alternative method of meniscal replacement is therefore needed.

The predominant orientation of the collagen fibre bundles within the meniscus is circumferential (Bullough et al 1970), with more radial fibres in the inner third (Beaupré et al 1981, 1986). Circumferential hoop stress develops during axial loading of the knee which enables the menisci to act as shock absorbers (Seedhom et al 1974; Kurosawa et al 1980). Histologically, the peripheral half of the meniscus is similar to tendon tissue (Fithian, Kelly and Mow 1990) and we hypothesised that a tendon of the appropriate size could be used to replace that part of the meniscus.

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We used the middle third of the patellar tendon as an autograft to see whether it would change in time to meniscus-like tissue and whether it could protect the articular cartilage from the degenerative changes which usually result from meniscectomy.

MATERIALS AND METHODS

Experimental protocol. We used 20 skeletally mature female merino sheep allocated randomly into four groups of five animals each. On the right hind leg we performed total medial meniscectomy, removal of the middle third of the patellar tendon, and tenotomy of the calcaneal tendon to ensure non-weight-bearing on the operated leg in the postoperative period. In group I (control group), no additional procedure was performed. In the other three groups (II, III and IV), the medial meniscus was replaced by the middle third of the patellar tendon of the ipsilateral knee. The joints were examined at three months in group II, six months in group III, and 12 months in groups I and IV.

Fig. 1

The middle third of the patellar tendon is introduced into the medial compartment of the knee to replace the peripheral part of the medial meniscus. Its anterior and posterior ends are fixed to the bone, and its periphery is sutured to the capsule.

Operative technique. After general anaesthesia (xylazine 0.05 mg/kg intramuscularly, ketamine 3 to 6 mg/kg intravenously and halothane/oxygen) the right hind leg was cleaned and draped. Through a medial parapatellar incision the medial collateral ligament was detached from the femur and the entire medial meniscus removed. The hyaline cartilage of the tibiofemoral joint was inspected. We removed part of the middle third of the patellar tendon corresponding to the peripheral length of the medial meniscus. No attempt was made to shape it to a triangular cross-section. Both ends were tagged with

non-absorbable threads (Ethibond) and the graft covered with polyglactin nettube of 8 mm diameter (Vicryl-Schlauch, Ethicon GmbH, Norderstedt, Germany). Holes, 2 mm in diameter, were drilled from the anterior surface of the tibia to the anterior and posterior areas of insertion of the meniscus (Fig. 1), as located by the remnants of the original meniscal attachments. The tag sutures were passed through the drill holes and the tendon was inserted into the medial joint space and sutured to the capsule with four absorbable Vicryl threads. The femoral condyle was repositioned and the medial collateral ligament reattached to it by two transosseous sutures. With the joint in 45° of flexion the tendon was pulled tightly around the condyle by the tag sutures which were then tied (Fig. 1). The wound was closed in layers and prophylactic antibiotics given routinely. Movement was not restricted in the postoperative period.

Evaluation. Postoperatively, wound healing and gait were examined twice weekly. After the animals had been killed at the appropriate interval, the right hind leg was disarticulated at the hip, stability of the knee was

Fig. 2

Fig. 3

Figure 2 - The retrieved autografts were sectioned for histology (H), scanning electron microscopy (SEM), and biomechanical tests (B). The anterior (A) and the posterior (P) fixation sites were not examined (MCL = medial collateral ligament). Figure 3 - The size and shape of the specimens used for biomechanical testing.

evaluated by manual testing, and radiographs of the joint obtained in two planes. The knee was then opened, assessed macroscopically, and photographed. The tendon autografts in groups II, III and IV and the regenerated tissue in the control group were removed and divided into specimens for examination by light and scanning electron microscopy and for biomechanical evaluation (Fig. 2).

Micro-angiography. At the end of each of the three intervals, one sheep in groups II, III and IV was again anaesthetised as described above. Heparin (20 000 IU) was given, the right femoral artery was cannulated and the right hind leg perfused with 500 ml of dextran 40 solution. After killing the sheep, the right hind leg was disarticulated at the hip and perfused with a radio-opaque suspension containing 300 ml of barium sulphate and 700 ml of Ringer-Lactate solution at a perfusion pressure of 200 cm H₂O for 12 hours. The joint was then opened and the tendon graft removed together with adjacent capsular tissue and frozen at -20°C. During the following 48 hours, soft radiographs of the specimens were made (Microplus NIF 100 film; DuPont de Nemours, Deutschland GmbH, Frankfurt, Germany). We performed a similar procedure on the left hind leg of one animal in group I to obtain a micro-angiogram of a normal medial meniscus for comparison.

Histology and scanning electron microscopy. In all animals not included in the micro-angiography series, the specimens of tendon autograft were immersion-fixed *en bloc* immediately after removal in 3% glutaraldehyde buffered with 0.1 M sodium cacodylate/HCl. Specimens taken from tissue close to the medial collateral ligament were used for light microscopy. They were rinsed in buffer, post-fixed for two hours in a 1% osmium tetroxide solution in distilled water, dehydrated in graded ethanols and embedded in Epon or in paraffin. Each block was cut into 1 µm thick sections and stained with either an alkalised toluidine blue solution, haematoxylin and eosin, or Van Gieson's stain. The larger posterior part of the specimens (Fig. 2) was used for scanning electron microscopy (SEM). After pre- and post-fixation as for light microscopy, they were dehydrated in a graded series of acetone, critical-point dried with carbon dioxide, mounted onto stubs with carbon and sputter-coated with gold-palladium. SEM was performed with accelerating voltages between 8 and 25 KV (Philips SEM 505; Philips GmbH, Hamburg, Germany).

Biomechanical analysis. We performed biomechanical studies on the anterior two-thirds of the specimens (Fig. 2). The five specimens of the middle third of the patellar tendon and the five medial menisci removed from the group I animals were used for comparison. The dissected grafts were stored at -20°C in airtight bags. Dumb-bell-shaped specimens 1 mm thick (Fig. 3) were removed by deep-freeze cutting. On the day of testing, they were thawed at room temperature and fixed with cyanoacrylate between the specially designed clamps of a tensiometer.

The free length of the specimen was 2 mm. We performed tensile tests at 20°C at a displacement rate of 3 mm/min using a uni-axial testing machine (Zwick 7025-3, FA ZWICK, Ulm/Donau, Germany). The specimens were kept moist throughout the test by isotonic NaCl solution. Before testing a pre-load of 0.1 N was applied for two minutes. Cross-head displacement and tensile force were recorded on load-deformation diagrams during testing to failure.

RESULTS

There were no intra- or postoperative complications. All the animals were lame on the operated hind leg for at least four weeks postoperatively and avoided weight-bearing on this leg. After 12 weeks, gait had returned to normal and no gap could be palpated in any calcaneal tendon.

Manual testing of stability after disarticulation of the leg showed no medial instability in any of the operated knees. Anteroposterior and lateral radiographs at 12 months revealed early degenerative changes with flattening of the medial condyle and narrowing of the joint space in six of the 20 animals, four in the control group (I) and in two in the meniscal replacement group (IV).

Macroscopic findings. At 12 months the meniscus had regenerated in all the control animals (group I), but none was more than 25% of the width and 50% of the height of a normal meniscus.

In six animals in groups III and IV the autograft was similar to the original meniscus in shape and size (Fig. 4) and was termed a complete tendon-meniscus. In eight animals, it was the same shape as a normal meniscus but smaller; this was called an incomplete tendon-meniscus. In only one animal did we find a small regenerated meniscus similar to that seen in the control group.

In the animals killed at three months (group II), five incomplete tendon-menisci were found. After six months (group III), there were three incomplete and one complete tendon-menisci, as well as the one regenerated meniscus. After 12 months (group IV), all animals had a complete tendon meniscus. One incomplete (group III) and one complete tendon meniscus (group IV) had bucket-handle tears. The fragments comprised about a quarter of the width of the menisci and were displaced centrally.

At the time of the operation no cartilage defects were seen in the medial tibiofemoral joint. Twelve months after meniscectomy, all animals in group I had fibrillation and discoloration of the medial tibial plateau and softening and discoloration of the cartilage on the medial femoral condyle. After meniscal replacement, three patterns of hyaline cartilage were seen. Intact hyaline cartilage was found in the five animals in group II. Intact cartilage on the femoral condyle and on the central area of the tibial plateau, with an indentation of cartilage beneath the tendon-meniscus, was found in

Fig. 4a

Fig. 4b

Figure 4a – A complete tendon-meniscus (TM) and a normal lateral meniscus (LM) (group IV). The sectioned stump of the anterior cruciate ligament (ACL) is seen. Figure 4b – Complete tendon-meniscus after removal. The arrow marks the anterior tag suture. (A = anterior.)

three animals in group IV (Fig. 5). In five animals in group I and in two in group IV, there was fibrillation of the centre of the tibial plateau and softening of the cartilage of the medial condyle without indentation beneath the tendon-meniscus.

Histology. Light microscopy of cross-sections of normal sheep menisci showed collagen fibre bundles predominantly of the circular type, and radiating fibres mostly confined to the femoral articular surface (Fig. 6). Chondrocyte-like cells were seen. Patellar tendon tissue had typical fibroblasts but no chondrocyte-like cells.

Three months after operation (group II), necrosis of the small vessels was detectable in the patellar tendon autografts. After six months (group III), highly cellular connective tissue had developed adjacent to the central (free) edge of the tendon (Fig. 7). It was triangular in cross-section and contained blood vessels. The peripheral tissue, near the capsule, and the newly formed connective tissue centrally, together created the 'tendon-meniscus'. The two parts of this structure could still be observed histologically in group IV specimens 12 months after operation – a peripheral part with strong circularly disposed collagen fibre bundles and a central part with fewer fibres and more cells (Fig. 8). These appearances suggest that a remodelling process had taken place (Fig. 9). As well as circular collagen fibre bundles, radial and oblique fibres were also present (Fig. 10). Chondrocyte-like cells were found within the tendon-meniscus tissue both centrally and peripherally (Fig. 11).

Micro-angiography. Micro-angiography showed complete vascularisation of the autografts in the three-month and six-month groups (II, III). After 12 months (group IV), the central one-third of the tendon-meniscus had become avascular but the peripheral two-thirds were still vascularised (Fig. 12a). This is in contrast to the normal sheep meniscus, which is vascularised only near its capsular attachments (Fig. 12b).

Biomechanics. The failure stress and the tensile modulus for the tendon-meniscus were significantly lower than for

Fig. 5

Tibial plateau after removal of a complete tendon-meniscus and the lateral meniscus (group IV). There is an indentation (arrowheads) beneath the tendon-meniscus and fibrillation of the hyaline cartilage in the centre of the tibial plateau (arrow).

the normal meniscus and the normal patellar tendon (Fig. 13). The highest values were obtained at the end of the observation period.

DISCUSSION

Some parts of the human body can be removed without ill effect when they are diseased or damaged; some parts regenerate completely. Neither of these statements is true of the meniscus of the human knee (Fairbank 1948; Tapper and Hoover 1969; Krause et al 1976). After meniscectomy, in man, the pressure on the articular cartilage is increased and the area of direct tibiofemoral contact is enlarged (Kettelkamp and Jacobs 1972). In sheep, chondromalacia of the medial tibial plateau regularly occurs after meniscectomy. The question remains whether a meniscal substitute can prevent degenerative changes in the meniscectomised knee.

The most important role of a meniscal substitute is

Fig. 6

Figure 6 - Normal sheep medial meniscus, femoral surface above, showing the collagenous fibre bundles (triangles) and chondrocytes (arrowheads) (Van Gieson $\times 14$). Figure 7 - Incomplete tendon-meniscus (group III). Its central part consists of highly cellular connective tissue with vessels (arrowheads) and fibres (Van Gieson $\times 23$).

Fig. 7

Fig. 8a

Fig. 8b

Scanning electron microscopy of a tendon-meniscus (group IV). Figure 8a - Central part showing fibre bundles (F). Inset shows blood vessels in the free margin of the meniscus (arrows). Figure 8b - Peripheral part showing the capsule (C). ($\times 16$; inset $\times 140$).

the protection of the hyaline cartilage and the best criterion by which to judge its success is the state of the cartilage of the corresponding plateau (Bullough et al 1970). The indentation beneath the tendon-menisci in three of our animals suggests that load was carried through the new meniscus. Neither the shape of the autograft at the time of implantation nor its material properties are ideally suited for its purpose. The cartilage beneath the implant will therefore deteriorate at least

during the first few months but it seems that this may protect the cartilage in the centre of the plateau.

Attempts to use a collagenous meniscal prosthesis (Stone et al 1990) have not yet led to a clinically applicable procedure. Replacement of the meniscus by a meniscus would seem to be the most logical approach and many investigators (Keene et al 1987; Milachowski et al 1988; Zukor et al 1990; DeBoer and Koudstaal 1991) have described attempts to replace the entire meniscus, but no

Fig. 9

Complete tendon-meniscus (group IV) showing fibrous connective tissue with strong circular bundles and chondrocyte-like cells (Van Gieson $\times 90$).

Fig. 11

Scanning electron micrograph showing chondrocyte-like cells in a complete tendon-meniscus (group IV) ($\times 750$).

Fig. 10

Complete tendon-meniscus (group IV) showing collagen fibre bundles in cross-section (Q), cut obliquely (O) and lying in the plane of the section (L) (toluidine blue $\times 300$).

Fig. 12a

Fig. 12b

Figure 12a - Angiographs of a complete tendon-meniscus (group IV) showing vessels extending throughout its peripheral two-thirds. Figure 12b - Angiograph of a normal medial meniscus. Only the capsular attachment is vascularised.

Fig. 13

Failure stress (solid circles) and tensile modulus (open circles) (mean $\pm$ SD) for normal medial meniscus tissue (GM), patellar tendon tissue (PT) and tendon-meniscus tissue (TM) three, six and twelve months after implantation. The stars mark significance on a 0.05 confidence level (U test).

safe and viable transplant has yet been found (Arnoczky and Milachowski 1990). Perhaps functional replacement of a meniscus could be achieved without anatomical reconstruction. Shock absorption could be provided by any semicircular structure fixed at both ends to the tibial plateau. Such a structure would be distended by the femoral condyle under load (Seedhom et al 1974; Shrive et al 1978). Its cross-sectional shape may be less important than the fact that it should be in contact with the femur on one side and the tibia on the other and it may be that the exact positioning of the anterior and posterior attachments of the meniscal substitute is as important as the isometric placement of a graft of the anterior cruciate ligament.

Histologically, tendon tissue is similar to the peripheral tissue of the meniscus (Bullough et al 1970; McDevitt and Webber 1990) although there are some distinct differences.

Fibrochondrocytes are not found within tendon (McDevitt and Webber 1990), and the fibre orientation in tendons is exclusively parallel whereas in the meniscus there are radial fibres and a densely woven surface layer (Beaupré et al 1981, 1986). As has been known for a long time, however, connective tissues can transform to fulfil new functions after transplantation if conditions are favourable (Roux 1895). The tensioning of the tag sutures after insertion of the tendon provided pre-load and may have been critical for the outcome of the tendon autograft procedure.

Some tendon-menisci in our study had become triangular on cross-section by six months. By 12 months the space formerly occupied by the meniscus was entirely filled and the tendon-menisci fitted the individual knee perfectly. For lubrication and nutrition of the hyaline cartilage, the filling of the space formerly occupied by the meniscus may be more important than the mechanical properties of the implant (MacConaill 1950). Individual shaping of the meniscus is not possible if a meniscal prosthesis is used (Stone et al 1990). The efficient transmission and distribution of pressure across the joint are possible only if the meniscal substitute is totally congruent with the femur and the tibia (Beaupré et al 1981, 1986). After initial necrosis, a remodelling process took place within our specimens which gave their tissues some of the qualities of meniscal tissue. Chondrocyte-like cells were seen after six months and the characteristic pattern of the fibre bundles was achieved after 12 months. Ghadially, Wedge and Lalonde (1986) reported that after meniscectomy synovial tissue filled the space that was created; we think that the newly formed tissue of the central part of the tendon-menisci probably had a synovial origin, filling the 'dead space' which the tendon could not fill.

The two bucket-handle tears in the tendon-menisci draw attention to the problem that tendon tissue may split because of the exclusively parallel direction of its collagen fibre bundles. If part of the tendon slips centrally

into the knee before a synovial layer has covered it, a bucket-handle tear will result.

The main angiographical and histological differences between genuine menisci and tendon-menisci were the presence of blood vessels and the different pattern of fibre bundles in the latter. Biomechanically, the differences between patellar tendon, normal meniscus and tendon-meniscus were considerable and cast doubt on the suitability of the tendon as an autograft. The changes which the tendon autografts underwent were similar to those which occur when they are used to replace ruptured cruciate ligaments (Bosch et al 1989). Our observations suggest that the tendon does not change into a new meniscus but only into a somewhat inferior substitute.

We cannot explain why three animals in the 12-month group had no chondromalacia in the centre of the tibial plateau and two had chondromalacia. Perhaps variations in the thickness of the tendon and its pre-loading at implantation played a role.

We propose that the tendon-meniscus be considered as one option for medial meniscus replacement in man in the future.

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