

In vivo high-resolution three-dimensional magnetic resonance imaging of a rat knee osteoarthritis model induced by meniscal transection

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Introduction: In a surgically induced model of knee osteoarthritis (OA), a full-thickness cut in the medial meniscus leads to joint instability and progressive development of OA characterized by proteoglycan loss, cartilage fibrillation, chondrocyte death, subchondral bone changes, and formation of osteophytes [1; 2]. This OA model offers the advantages of site specificity, controlled disease onset and a relatively short development time [1]. Using this model it has been demonstrated that MMP inhibition is effective in reducing the joint damage [2], and fibroblast growth factor-18 can stimulate repair of damaged cartilage [2; 3]. The current study was aimed to evaluate in vivo high-resolution 3D MRI in demonstrating the joint structural changes in this OA model in rats.

Materials and methods:

Meniscal transection was carried out on the medial aspect of the left knee of male Lewis rats (n=10, 200-250g). Sham-operated animals (n=5) underwent surgery however transection of the meniscus was omitted. MR scan was carried out 44 days after surgery using a 4.7T Varian system equipped with a 300mT/m gradient. The radio frequency coil was an in-house built double balanced matched 3cm diameter copper sheet solenoid, with 1cm in length. A 3D data set was acquired using a spoiled multi-echo fat-suppressed 3D gradient echo (TR=75ms, flip angle=30°, 5 echoes TE1=2.8ms, TE2=6.0ms, TE3=9.2ms, TE4=12.5ms, TE5=15.7ms). Echo summation provided a means of enhancing SNR and enabled us to acquire high resolution 3D image of the rat knee in approximately 50mins. The images covered the entire knee joint with a resolution of 59*117*234µm. Image reconstruction was performed using in-house software developed in Matlab. A modified version of ImageJ [National Institute of Mental Health, USA] was used for image analysis. After MRI scan, the joints were processed to Toluidine blue stained sections and haematoxylin-eosin stained when necessary. Procedures were performed in full compliance with licenses issued under the UK Animals (Scientific Procedures) act, 1986.

Results:

Medial meniscus transection was clearly visible on MR images post surgery. The anterior half of the meniscus retracted anteriorly and the posterior half retracted posteriorly. Menisci of sham operated animals were unaffected. Of the ten animals with meniscal transection, cartilage damage with varying degree of severities was observed in all animals, including decrease of cartilage thickness and loss of cartilage in some areas, and focal neo-cartilage proliferation at joint margin. Subchondral bone change was identified in 9 animals with surgery, again with varying severities. Damage to the subchondral bone included local osteosclerosis and deformed tibia cortex surface. Osteophytes occurred on the medial joint margin of the tibia plateau in some animals. Cartilage and bone degeneration were most extensive on the weight-bearing region of the medial tibial plateau. No apparent subchondral bone damage was observed in the epiphysis of the femur. In five animals single or multiple high signal areas were seen within the epiphysis of tibia, consistent with cyst signal. The inter-articular space on the medial side was slightly increased in two animals with surgery. Mild femur-tibia axis misalignment was seen in one animal. Bone and cartilage anatomy was normal in all sham-operated animals.

Histopathological changes were characterised by a variable severity of cartilage degeneration. There was loss of chondrocytes, and proteoglycan evidenced by reduced toluidine blue staining. Fibrillation of the superficial cartilage, vertical and lateral fractures in the cartilage matrix and ulceration generally to the tide mark were common findings. Notable chondrocytes were present at the lateral borders of both the medial tibial and femoral condyles. These histopathological changes were generally most severe in the medial tibial cartilage. In some joints further degeneration and destruction of subchondral bone occurred in areas of cartilage degeneration or ulceration. In these locations extrusion of degenerative articular cartilage and some bone spicules into the underlying epiphyseal marrow spaces was often seen. Within these areas there was a loss of the normal marrow with replacement by cellular infiltrates which included activated osteoclasts, osteoblasts and fibroblast-like cells with fibrous matrix formation. These areas occasionally replaced a substantial proportion of the marrow space and with further degeneration of the cancellous bone of the epiphysis, formed sizable cysts. Occasionally the cystic areas were relatively acellular and contained eosinophilic material, presumably fluid. It was notable that epiphyseal cysts were only present when there was evidence of subchondral bone destruction. These epiphyseal cysts were consistent with the high signal areas seen in the epiphysis of tibia by MRI.

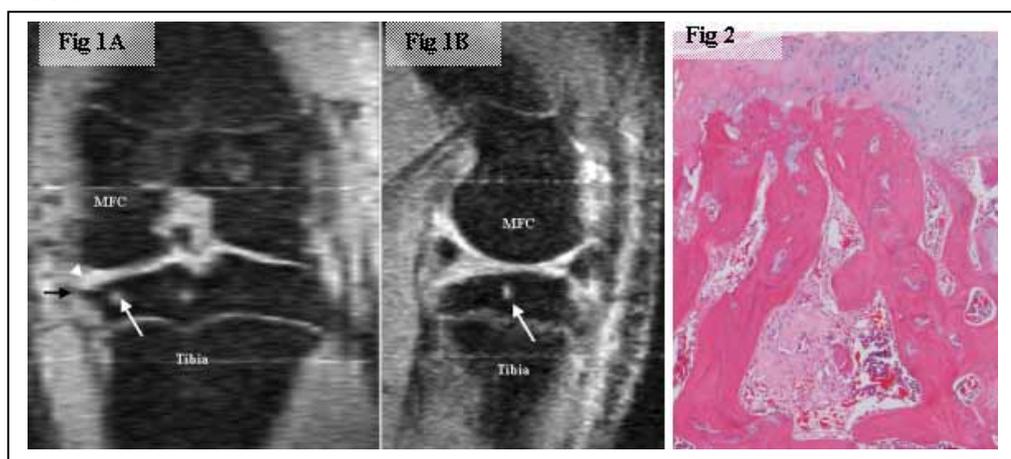


Fig1: MRI of rat knee OA with medial meniscal transection. A: coronal view; B: sagittal view. Fig1A shows the location of meniscal transection indicated by a white arrowhead. Bone damage occurred at the medial 1/3 of the medial tibia plateau. Peripheral osteophyte is indicated by a black arrow. A cyst signal is seen in the epiphysis of tibia (white arrow). MFC: medial femur condyle.

Fig2: The figure illustrates localised degeneration of the tibial articular cartilage and subchondral bone following meniscal transection. There is extrusion of degenerative articular cartilage through the subchondral bone plate into the underlying epiphyseal marrow space. Within these areas there is a loss of the

normal marrow with replacement by cellular infiltrates including mononuclear inflammatory cells and activated osteoclasts, and fibrous tissue formation.

Discussion:

To our knowledge, this is the first report using in vivo MRI to assess rat knee OA induced by meniscal transection. Due to nature of pathology in this model, very high resolution is needed to demonstrate the structural changes. Our results demonstrated that MRI was able to characterise knee OA induced by meniscal transection, and changes seen on MRI were consistent with histopathological data. A particular interesting finding from this study was the cyst formation in tibia epiphysis. To our knowledge, this has not been reported with this OA model. Pain is the principal symptom of OA [4]. Rat knee OA induced by meniscal transection has been used a joint pain model [5]. It is well known that cartilage does not contain pain fibers. Bone in the periosteum and bone marrow is richly innervated with nociceptive fibers. The association between these bone cysts and pain warrants further investigation.

References: 1. Bendele A, et al *Toxicol Pathol* 1999;27:134-42. 2. Janusz MJ, et al. *Osteoarthritis Cartilage* 2002;10:785-91 3. Moore EE, et al. *OsteoArthritis and Cartilage* 2005; 13, 623-631 4. Dieppe PA et al. *Lancet*. 2005;365:965-73 5. Fernihough J, et al. *Pain*. 2004;112:83-93