How do Bone Marrow Lesions Cause Osteoarthritis Pain? A structural and functional tissue-based study

N Sofat, L Assi, A Kuttapitiya, A Boyde, V Ejindu, C Heron, FA Howe

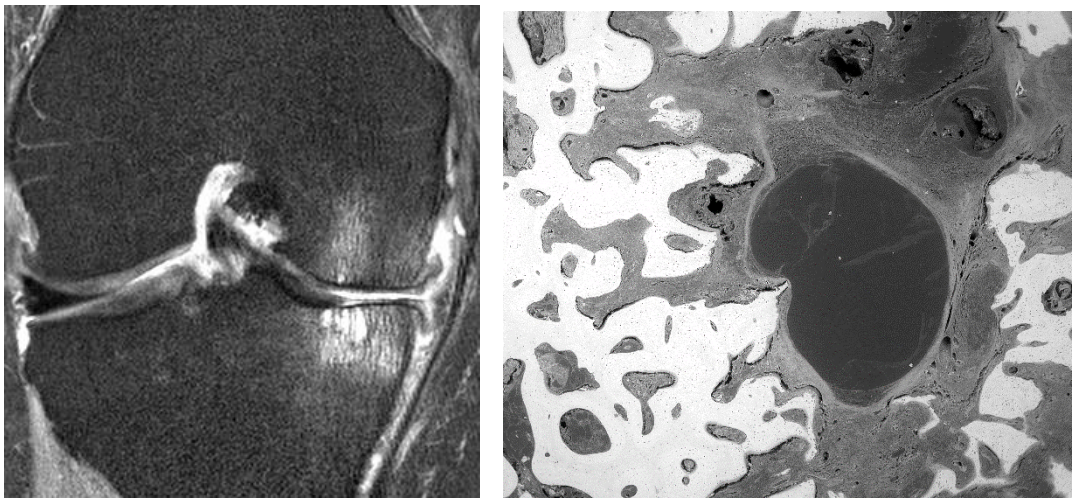
Background/Purpose:

Bone marrow lesions (BML) are well described in osteoarthritis (OA) and associate with pain. However, little is known about histological and functional features of BMLs. Our primary aim was to evaluate BMLs using novel tissue analysis tools to gain a deeper understanding of how they mediate pain.

Methods: We recruited 96 participants into the study. Participants with ACR criteria for knee OA were recruited (n=84), who were divided into advanced OA (n=72), who had severe enough disease to require total knee replacement (TKR). Early knee OA subjects had pain but did not require surgery (n=12). An additional 12 controls were recruited to control for pain measures and tissue comparisons from participants undergoing surgery for non-OA reasons. All participants were assessed by Western Ontario and MacMaster Universities Osteoarthritis Index (WOMAC). All subjects had knee MRI to define BML characteristics, synovitis and cartilage damage, scored using the MRI Knee Osteoarthritis Score (MOAKS). Tissue was harvested at TKR for BML analysis using scanning electron microscopy (SEM) and tissue microarray using Illumina. For SEM, tissue blocks were embedded in poly(methyl methacrylate) to give intact tissue and analysed to obtain 3 Dimensional SEM. For microarray, RNA was isolated and reverse transcribed using the Qiagen system, then subjected to microarray using standard Illumina protocols.

Results: The mean (SD) total WOMAC scores in the groups were as follows: advanced OA 1436.2 (471.6), early OA 797.4 (549.6) and controls 10.5 (12.6), demonstrating that the advanced OA group had severe pain ($p<0.0001$).

Figure 1. A. MRI showing BML in femur and tibia B. SEM of BML showing cyst and infiltration with cartilage



MOAKS scoring in the advanced OA group showed 90.1% had femoral and tibial BMLs. SEM showed most normal bone marrow was adipocytic with adipocytes the major bone lining cells, often making and moulding trabecular excrescences. Bone volume fraction was starkly reduced in BML areas, with marrow replaced by dense fibrous connective tissue, hyaline cartilage and fibrocartilage. Areas of aggressive resorption were found at the periphery of BML patches and areas of calcified cartilage so deep within the bone that they could not be explained by impaction from the joint surfaces, but were arising by mineralisation of cartilage formed deep within the bone organ (Figure 1). Tissue microarray of a subset of n=24 samples from the OA BML and controls showed 218 genes were significantly differentially regulated compared with control samples ($p<0.05$). Gene groups demonstrating the highest levels of regulation included the extracellular matrix proteins, the chondrocyte-expressed protease matrix metalloproteinases (MMP-13), neuro-epithelial and axonal development proteins, pro-inflammatory cytokines and catenin signalling.

Discussion: Our study is the first to employ SEM and microarray techniques in one study to interrogate OA BMLs. We have found that BMLs demonstrate areas of high metabolic activity, comprising of cartilage proteins and enzymes in addition to neuronal differentiation proteins which could explain why they are strongly associated with pain.