MRI Assessment of Temporal Soft Tissue and Bone Changes in Murine Collagen-Induced Arthritis

A. Pohlmann[1,2], V. Musoko[2], O. Woolmer[3], A. Robinson[3], J. Buckton[4], A. R. Hobson[4], and M. F. James[2]

[1]Academic DPU, GlaxoSmithKline R&D Ltd, Cambridge, United Kingdom, [2]Immuno-Inflammation CEDD, GlaxoSmithKline R&D Ltd, Harlow, United Kingdom, [3]Laboratory Animal Science, GlaxoSmithKline R&D Ltd, Harlow, United Kingdom, [4]Immuno-Inflammation CEDD, GlaxoSmithKline R&D Ltd, Stevenage, United Kingdom

Introduction: In collagen-induced arthritis (CIA) in mice the knee joints are preferred for imaging arthritic change [1], although the paws are used to assess disease severity and therapeutic efficacy. We developed in vivo MRI of hindpaw changes for direct comparison with conventional scoring procedures and studied the temporal progression of CIA.

Materials and Methods: Animal experiments complied fully with UK ethical and legal requirements. We studied the hind paws of 20 DBA mice: 6 naïve and 14 that received collagen (100µg bovine type-II, Freund's Complete Adjuvant) injected into the tail base on Day -21 and a second booster injection of collagen i.p. on Day 0. Arthritis severity was scored visually (0-4 for each paw) 3x/week until Day 0, and every weekday thereafter. From Day 0 hindpaw thickness was measured with digital calipers. The hindpaws were imaged first between Day 2 and Day 10 (in order of arthritis severity) and then weekly for 4 weeks. *Imaging.* 25 T1-weighted axial images were acquired in 28 minutes on a 4.7T Bruker Biospec (FLASH, TR/TE/FA = 521ms/2.4ms/50°, FOV = 23×11.5×17.5mm, resolution 0.09×0.09×0.5mm, fat-suppressed). Contrast agent (Omniscan, 0.1mmol/kg) was injected i.v. after image 15. *Analysis.* The T1w data were analysed using principal component analysis, producing an average image (eigenimage-1) with high SNR and a map of contrast agent-induced signal change (eigenimage-2). Both images were segmented using a threshold of 4 standard deviations above mean background noise, yielding the cross-sectional area of soft tissue or enhancing tissue per image slice respectively. To allow averaging of the area-vs-length profiles (Fig.1) we aligned corresponding slices between paws using anatomical landmarks. Area-under-curve calculations gave corresponding volumes. The volume of 'cavities' within the segmented soft tissue was interpreted as bone/cartilage. The outline of the segmented bones was used as an indicator of bone surface changes (expected during bone re-modelling).

Results and Discussion: Plots of tissue area show how soft tissue swelling develops along the hindpaw length (Fig.1), reflecting the arthritis scores (Fig.3). However low scores (<1) reflected MRI changes poorly indicating the relative inaccuracy of visual assessment. Volumes of enhancing tissue (Fig.2) reflect hindpaw thickness measurements (Fig. 3), increasing progressively with time, although not quite matching the soft tissue changes (Fig.2). Score, paw thickness, soft tissue and enhancing tissue volume were strongly correlated (Table 1). Bone volume or outline did not change significantly (Fig.4), but a weak trend towards decreasing volume and increasing outline was observed, which agrees with the expected trends during bone degradation. The sensitivity and accuracy of the bone analysis may have been insufficient to detect changes within this short time period. Data variability may have been increased by inaccuracies in the image slice alignment or automated segmentation; in sections where bone was covered only by a thin layer of skin the soft tissue cavities (representing bone) required manual 'closing' to calculate their volume.
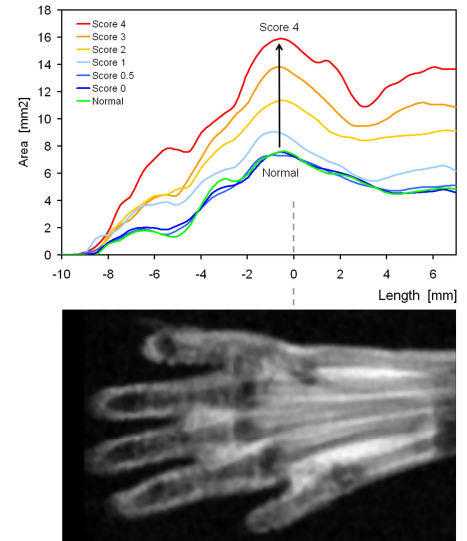

Fig.1: *Above*: Plots of mean cross-sectional area of soft tissue vs length for animals with different scores. *Below*: A corresponding coronal MRI image.

Table 1: Correlation coefficients

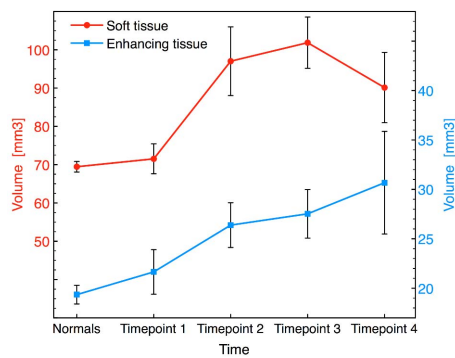|	Soft Tissue	0.69	Enh. Tissue
Score	0.85		0.62
0.62			
Thickness	0.72		0.60

Fig.2: Volume (mean±SEM) of soft tissue and enhancing tissue for normal paws and arthritic paws at four timepoints.
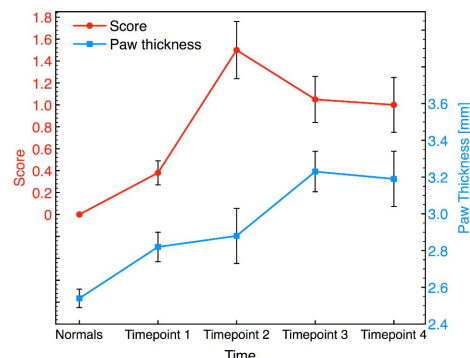
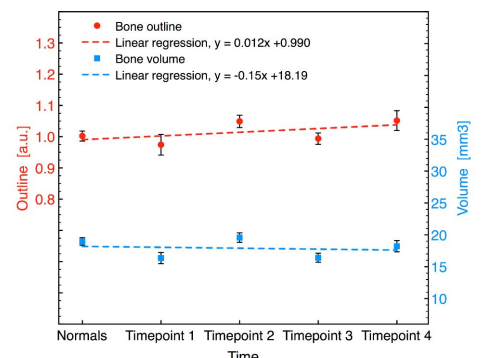**Fig.3:** Score and paw thickness (mean±SEM) for normal paws and arthritic paws at four timepoints.

Fig.4: Bone volume and outline (mean±SEM) for normal paws and arthritic paws at four timepoints. Linear regression shown as dashed lines.

Conclusions: Our results show MRI detects soft tissue and perfusion changes in hindpaw arthritis sensitively and objectively and in a quantitative manner. Whereas scoring represents an integrated assessment or arthritic changes, imaging may separate the underlying disease processes (swelling, perfusion, etc). These results encourage further work to improve the bone analysis in order to be able to quantify changes observable in the skeletal elements more sensitively, so that a complete in vivo picture of the progressive inflammatory, perfusion and skeletal changes can be defined.

References: [1] Proulx, ST, 2007, Arthritis Rheum 56(12), 4024-37.