Chronic axial compression of the mouse tail segment induces MRI bone marrow edema changes that correlate with increased marrow vasculature and cellularity

**INTRODUCTION:** Although MRI evidence of bone marrow edema (BME) in vertebral adjacent to degenerative discs remains a common diagnostic finding in clinical scans performed on patients with back pain associated with degenerative disk disease (DDD) and early spondyloarthopathy (SA), our ability to interpret this information is limited by a lack of knowledge of its cellular nature and natural history. To address this, we have developed a mouse model utilizing chronically loaded tail vertebrae that closely resembles the radiology and histopathology of early vertebral degeneration in humans. We hypothesize that these changes are caused by the conversion of marrow from a fatty "white" marrow to a hematopoietic "red" marrow including the infiltration of the endosteal surface of the affected vertebral endplates.

**METHODS:** Animals: Wild type C57bl/6 (WT) heterozygous TNF transgenic mice maintained in C57bl/6 background (TNF-Tg). Animals: Two 0.28" diameter titanium pins were implanted transcutaneously in the center of the 7th and 10th caudal vertebrae (Fig. 1A). Aluminum rings were attached to the implanted pins, which allow for chronic loading via manual tightening of four screws around which calibrated springs were placed (Fig. 1B). A load of 6X body weight was applied over an 8 week trial along with age matched unloaded controls. Mice were euthanized at eight weeks and the compressed vertebrae were harvested, fixed, and processed for histological examination.

**Imaging:** Mice were anesthetized prior to MRI scans with a mixture of ketamine 60mg/kg and xylazine 4mg/kg injected I.P. to ensure minimal motion during scans. All instrumentation was removed prior to CE-MRI. We used custom built a small animal RF surface coil placed about the tail (Fig. 1C) that interface with a clinical 3 Tesla Siemens Trio MRI. A fat-suppressed, T1-weighted high-resolution scan was then performed as previously described. To assess changes in the marrow space due to bone marrow edema (BME) an indication of degeneration, Gd-DTPA contrast agent (Omniscan, Amersham Health, Oslo, Norway) is diluted for CE-MRI (Fig. 1D). Representative AB/OG stained histological sections (A-F) of the 8th and 9th caudal vertebrae from the 0X loaded (A,B) and 6X loaded (C-F) WT (A,D), and 6X loaded TNF-Tg (E,F) mice 8 weeks after implantation are shown at 10 X and 40 X magnifications. Of note is the ruptured annulus and hypertrophic nucleus pulposus of the herniated intervertebral disk in the 6X loaded (C) vs. 0X (A) loaded WT, which occurs in the absence of detectable synovitis vs. TNF-Tg (E,F), synovitis indicated by arrows. Histomorphometry demonstrated that the 6X load significantly increased the vascular sinus space (G), and the cellularity (H), of WT marrow (*p<0.01).

Collectively, these data demonstrate that chronic load-induced DDD causes similar BME signals in vertebral endplates to that observed in ankylosing spondylitis (AS), and correlates with a conversion from yellow to red marrow, with increased vascularity.

**DISCUSSION:** Similar bone marrow responses have been shown to be directly related to an inflammatory response mediated by TNF. Likewise TNF has been shown to play a role in adipose lysis and proliferation and activation of TNF-producing macrophages. Off label use of anti-TNF therapy have shown some promise in treatment of debilitating chronic back pain. While the immune suppression of such treatments may offset the benefits of slowing degenerative changes, understanding the transduction pathway from mechanical stress to chronic degeneration may allow us to intervene at some less immune compromising point.

**REFERENCES**

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