**THE CATHEPSIN K INHIBITOR L-006235 DEMONSTRATES BOTH DISEASE MODIFICATION AND ATTENUATION OF PAIN BEHAVIOUR IN THE IN THE MIA MODEL OF OSTEOARTHRITIS**

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**BACKGROUND**

Cathepsin K (CatK) is a cysteine protease predominantly expressed in the oesoteclast and involved in bone resorption (1). CatK expression is up-regulated in human osteoarthritis (OA) synovium (1) and CatK mRNA expression is increased in human OA bone (1). CatK over-expression in mice leads to spontaneous synovitis and cartilage degeneration (2). Inhibitors of CatK attenuate lesions and biomarkers of osteoclastogenesis (3,4). CatK inhibition reduces mechnosensitivity of knee afferent nerve activity in a guinea pig model of spontaneous OA, thus suggesting a role for CatK in joint nociception during disease progression (5).

**AIM**

To investigate the effects of a selective CatK inhibitor, L-006235 (6), on both pain behaviour and joint pathology in an OA model in the rat.

**METHODS**

Male Sprague Dawley rats received intra-articular injection of saline or monosodium iodoacetate (MIA; 1 mg) into the left knee joint. One day before injection, rats were dosed with either vehicle or 30 mg/kg L-006235 (p.o. twice daily) and then every day for 28 days. Pain behaviour (weight bearing on the hind limbs and changes in distal hind paw withdrawal thresholds) was quantified before and after intra-articular injection of saline or MIA and at several time points during the 28 day treatment period. Rats were then euthanized and tissues taken for ex vivo analysis. Tibiofemoral joints were removed and post-fixed in neutral buffered formalin (4% formaldehyde), decalcified in EDTA, processed and scored (7). Haematoxylin and eosiin staining was conducted. Cartilage surface integrity was scored from 0 (normal) to 5 (full-thickness degeneration), and a total joint damage score (range 0–15) was calculated as cartilage surface integrity × length of cartilage involved in thirds. Inflammation was graded on a scale from 0 (lining cell layer 1–2 cells thick) to 3 (lining cell layer >9 cells thick and/or severe increase in cellularity). Osteophyte scores ranged from 0 (no osteophyte) to 3 (osteophyte >160 μm). Sections from the posterior half of the knee joints were dewaxed and re-calculated with calcium chloride and magnesium chloride, before tartrate-resistant acid phosphatase (TRAP5b) staining was conducted using a commercially available kit (F386A, Sigma-Aldrich, Dorset, UK). TRAP positive osteoclasts were quantified as previously described (7), under 40 times magnification from one end of the growth plate to the other end using the following criteria; 1) displayed purplish to dark red cytosol, 2) number of nuclei >3/oocyteast, 3) located within the subchondral bone area, comprising the area between the cartilage/bone junction and the growth plate. All behaviour and histology experiments were conducted in a blinded fashion. All data are presented as Mean ± SEM.

**RESULTS**

L-006235 prevented the development of MIA-induced weight bearing asymmetry and lowering of ipsilateral hindpaw withdrawal thresholds. Effects of L-006235 were not immediate and only significant from day 14 post model induction, indicating that L-006235 does not alter early inflammatory pain associated with the MIA model but rather prevents the development of OA-like pain behaviour. At the end of the study, intra-articular injection of MIA was associated with a significant increase in cartilage damage score, which was significantly reduced by L-006235. Furthermore, MIA-induced increase in osteophyte score was significantly reduced by L-006235, and there was a non-significant decrease in synovitis. L-006235 did not alter the number of TRAP5-positive multinucleated osteoclasts in MIA rats.

**CONCLUSIONS**

The CatK inhibitor L-006235 reduced MIA-induced development of pain behaviour and associated cartilage damage and osteophytes. Future studies will evaluate whether L-006235 can reverse established OA-like pain and joint pathology, thereby evaluating the potential of this drug/class of drugs to not only treat OA pain but also to slow the development of OA joint damage and thereby extend the time to total joint replacement.

**REFERENCES**