TRANSIENT RECEPTOR POTENTIAL CANONICAL CHANNEL 1 DEPENDENT PATHWAYS ARE REQUIRED FOR OSTEOCLAST FUSION AND MEDIATE OSTEOPOROTIC BONE LOSS

Umlauf D,¹ Hidding H,¹ Lindemann O,² Dankbar B,¹ Frank S,¹ Cromme C,¹ Dietrich A,³ Marshall R P,⁴ Amling M,⁴ Steiner M,⁵ Kornak U,⁵ Schwab A,² Pap T,¹ Bertrand J¹ ¹Institute of Experimental Musculoskeletal Medicine, University of Münster, Münster, Germany; ²Institute of Physiology II, University of Münster, Münster, Germany; ³Institut für Pharmakologie und Toxikologie, Philipps-University Marburg, Marburg, Germany; ⁴Department of Osteology and Biomechanics, University Medical Center Hamburg-Eppendorf, Hamburg, Germany; ⁵Institute of medical Genetics and Human Genetics, Charité – Universitätsmedizin Berlin, Berlin, Germany

10.1136/annrheumdis-2011-201237.16

The formation of osteoclasts involves a sequence of cellular events including monocyte migration towards each other, cell-cell contacts and fusion to multinucleated cells. Given that TRPC1 has a pivotal role in these specific processes in other cell types, the authors investigated the role of TRPC1 in osteoclastogenesis and studied the skeletal phenotype of TRPC1^{-/-} mice under physiological conditions as well as using an animal model of postmenopausal osteoporosis.

For all in vitro experiments, bone marrow macrophages were isolated from TRPC1^{-/-} mice and corresponding wild type (WT) controls and were cultured in the presence of macrophage colony-stimulating factor and receptor activator of nuclear factor κ -B ligand. Osteoclast-like cells were characterised by staining for tartrate-resistant acid phosphatase (TRAP). Using quantitative real-time PCR mRNA levels of TRPC1 and NFATC1 were analysed. Time-lapse videomicroscopy was used to study osteoclast migration and fusion. The intracellular Ca²⁺ concentration of osteoclasts was measured ratiometrically with the fluorescent Ca²⁺ dye Fura-2. The skeletal phenotype of 16-week old mice was investigated by μ CT-analyses of trabecular bone in the lumbar spine. Ovariectomy was performed on 12-week old sex- and aged-matched littermates as a model for human postmenopausal bone loss.

PCR analyses indicate that TRPC1 is hardly expressed in monocytes and preosteoclasts but gets upregulated during osteoclast-differentiation. In osteoclast formation assays, the loss of TRPC1 leads to impaired osteoclast differentiation with a 75% reduction in large osteoclasts. While μ CT analysis revealed no differences in bone phenotype of TRPC1^{-/-} mice compared to WT mice under physiological conditions, there were significant effects in the ovariectomy model of estrogen-deficiency mediated bone loss. In this model, TRPC1^{-/-} mice exhibited a reduced loss of trabecular bone volume (-28.9% in WT compared to -13.1% in TRPC1^{-/-}) and BTM (-3.2% in WT compared to +0.9% in TRPC1^{-/-}) in LW5. Analysing the underlying signaling pathways, the authors found no differences in Ca²⁺-oscillations and store-operated calcium entries and no differences in NFATC1 expression. In time-lapse microscopy, however, the authors observed a reduced capacity of TRPC1^{-/-} osteoclast precursors to migrate and to fuse, due to a reduced velocity, translocation and a reduced increase of cell area over time.

These results indicate that TRPC1 dependent pathways contribute to the migration and fusion of osteoclasts and that the loss of TRPC1 while not affecting physiological bone turnover has a clear effect on accelerated bone loss as seen in estrogen deficiency. Therefore, these data suggest that TRPC1 may be a new therapeutic target for rapid osteoporotic bone loss.