Results: There were 43 members from 30 local associations in Oulu, Mikkeli, Helsinki and Turku in these geocaching trainings. All these members are suffering from rheumatic and other musculoskeletal diseases (RMDs) and work as volunteers for sport and physical activity, and now for geocaching. By 12 october 2018 there were 48 reumageocaches made by 13 local associations. The caches were logged 6593 times. So local associations earned 173-4303 euros each to be used for promoting sport and physical activity programmes and active lifestyles.
Conclusion: Promoting an active lifestyle is part of the health and wellbeing policy in The Finnish Rheumatism Association. The goal is to encourage people of all ages to stay healthy by getting enough exercise.
The geocaching project shows that it is easy to some of local associations to start projects like this. Some associations thought that it is too difficult to learn new methods especially when their members are elderly people. Geocaching is increasingly popular, inclusive, a fun and healthy pastime for individuals of all ages. It is also great for groups like local associatons, families, friends, and youth groups working as teams. Those local associations that were involved in this project received a great budget for their local sport and physical activity programmes. It is great to see something evolving from the beginning and come into being.
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Cartilage, synovium and bone

OP0071 REVEALING THE LINK BETWEEN OSTEOARTHRITIS DEVELOPMENT AND MESENCHYMAL STEM CELL SENESCENCE

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Background: Tissue accumulation of $p16^{INK4A}$-positive senescent cells is associated with age-related disorders such as osteoarthritis (OA). These senescent cells induce a tissue loss of function through a particular secretory phenotype called SASP (senescence-associated secretory phenotype).
Objectives: Links between OA onset and cellular senescence remain poorly detailed. We wanted to determine the localization of articular senescent cells in *in vivo* OA mouse models and study the involvement of mesenchymal stem cells (MSC) senescence in OA pathogenesis.
Methods: Wild-type mice C57BL/6, SAMP8/R1 (senescence accelerated mouse-prone and resistant), transgenic $p16^{INK4A\ +/Luc}$ and $p16^{INK4A\ Luc/Luc}$ were used. Experimental OA was induced by intraarticular injections of collagenase (CIOA). Cartilage, synovial tissue and subchondral bone were analyzed by histology, RT-qPCR and micro-tomography. MSCs come from healthy human donors and primary chondrocytes from OA patients.
Results: (1) CIOA was induced in senescence-driven luciferase transgenic mice. Under CCD camera, a peak in luminescence was detected at day 24 post-injection revealing the presence of senescent cells in the joint. Remarkably, articular senescence is not only a marker of the pathology but contributes to OA onset: mice deficient in $p16^{INK4a}$, a main senescence-driving known cell cycle inhibitor, were partially protected against CIOA. These results were confirmed in C57Bl/6 mice after CIOA by showing an increase in gene expression for senescence, catabolic and inflammatory markers in the synovial tissue preceding cartilage degradation.
(2) MSCs found in synovial, cartilage, fat pad and bone marrow participate in joint homeostasis. Because MSC are proposed to be at the root of OA development, we hypothesize that cellular senescence onset in these progenitor cells would be a possible etiological factor for OA. We have established an *in vitro* $p16^{INK4A}$-induced senescence model on human primary MSC: their intrinsic properties such as self-renewing are altered during senescence onset. Furthermore, in co-culture conditions with chondrocytes from OA patients, senescent MSC lost their extrinsic chondroprotective properties.
(3) To *in vivo* challenge these findings, we rely on the mouse model of accelerated senescence SAMP8, which develop spontaneous OA at the age of 6 months with cartilage degradation, synovial hypertrophy, osteophytosis and subchondral bone remodeling associated to meniscal calcification. Isolated MSC from these mice express senescence but non-inflammatory markers ($p16^{INK4a}$, p21waf1, MMP13, TGF-β1). Remarkably, intra-articular injection of these isolated SAMP8-derived MSC compared to SAMR1-derived control MSC, in young wid-type C57Bl/6 mice, was suffisant by its own, to induce significant articular cartilage degradation (OA score of 12.2 ± 1.5 vs 6.1 ± 3.5 for SAMP8 and SAMR1 MSC respectively. p <0.05).
Conclusion: $p16^{INK4A}$-induced cellular senescence in MSC played a causative role in cartilage loss of function and OA pathogeny. *In vitro*, senescent MSC show altered intrinsic and extrinsic supportive tissue functions. *In vivo*, intra-articular injection of senescent MSC was sufficient to induce cartilage degradation. Specific targeting of such deleterious senescent cells could be an innovating and promising treatment in OA.
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OP0072 INHIBITION OF CLK2 AND DYRK1A BY SM04690 AS A NOVEL MOLECULAR REGULATOR OF WNT SIGNALING, CHONDROGENESIS, AND INFLAMMATION, A POTENTIAL DISEASE-MODIFYING TREATMENT FOR KNEE OSTEOARTHRITIS

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Background: In the synovial joint, Wnt pathway upregulation contributes to osteoarthritis (OA) by increasing osteocyte differentiation, cartilage thinning, and inflammation. SM04690, a novel small molecule, has previously demonstrated potential OA disease-modifying effects through Wnt pathway inhibition *in vitro* and *in vivo*.
Objectives: To elucidate the novel mechanism of action for SM04690 on Wnt pathway inhibition, chondrocyte differentiation, and anti-inflammation.
Methods: Wnt pathway activity was measured using a cell-based TCF/LEF luciferase reporter in SW480 colon cancer cells. A kinome screen (318 kinases) was performed. The effects of SM04690 on protein phosphorylation of serine and arginine rich splicing factors (SRSF proteins), Sirt1, and FoxO1 in hMSCs, chondrocytes, and synovial fibroblasts were measured by Western blot. The effects of SM04690 and siRNA knockdown (KD) on chondrogenic and Wnt pathway gene expression were measured by NanoString gene expression panels and effects on LPS-induced inflammatory cytokines (IL-6, IL-8, TNF-α) in BEAS-2B cells were measured by qPCR and ELISA. *In vivo*, the pharmacodynamic effects of SM04690 were evaluated in monosodium iodoacetate injection-induced and anterior cruciate ligament transection with partial medial meniscectomy rat knee OA models in which a single intra-articular SM04690 (0.1 μg, 0.3 μg, 1.0 μg) or vehicle injection was administered. Cartilage was isolated at Day 10 and 35; phosphorylation and expression of SRSF proteins, Sirt1, FoxO1, STAT3, and NF-κB were measured by Western blot.
Results: SM04690 was a potent (EC_{50}=11nM) inhibitor of Wnt signaling. Cdc-like kinases (CLKs) and dual-specificity tyrosine kinase (DYRK1A) were identified as molecular targets of SM04690. In hMSCs and chondrocytes, compared to DMSO, SM04690 potently inhibited CLK-mediated phosphorylation of SRSF proteins. SM04690 also inhibited DYRK1A-mediated Sirt1 and FoxO1 phosphorylation, thus increasing total and nuclear FoxO1 levels. Compared to siRNA control, DYRK1A/CLK2 dual KD increased expression of chondrogenic genes (*COL2A1, ACAN, COMP, CD44* [all P<0.05]). CLK2 and DYRK1A KDs each inhibited Wnt pathway genes (*AXIN2, TCF7, TCF4, LRP5, FZD6, FZD7, PITX2* [all P<0.05]) with no effects on β-catenin levels, compared to siRNA control. In synovial fibroblasts, compared to DMSO, SM04690 decreased phosphorylation of NF-kB and STAT3. In BEAS-2B cells, compared to siRNA control, DYRK1A KD inhibited inflammatory cytokine production (IL-6, IL-8, TNF-α [all P<0.05]), while DYRK1A/CLK2 dual KD enhanced anti-inflammatory effects of DYRK1A KD. In cartilage from rat OA models, compared to vehicle, SM04690 inhibited phosphorylation of SRSF proteins, Sirt1, FoxO1, and STAT3, as well as expression of NF-κB.

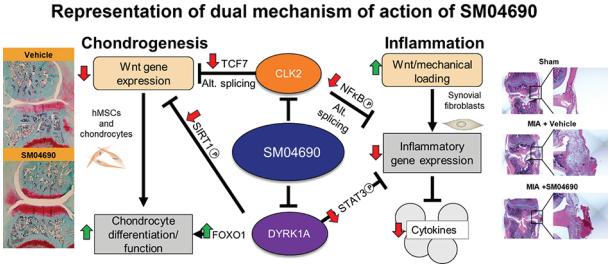

Figure 1. Schematic representation of SM04690's proposed mechanism of action via CLK2 and DYRK1A inhibition in OA

Conclusion: To our knowledge, this is the first report of SM04690 inhibition of nuclear kinases CLK2 and DYRK1A, leading to effects on the Wnt pathway, chondrocytes, and inflammation **(Figure 1)**. This dual mechanism of SM04690 potentially modifies OA through increased chondrocyte differentiation and function and benefits symptoms through anti-inflammatory activity. Human trials are ongoing.

Disclosure of Interests: Vishal Deshmukh Shareholder of: Samumed, LLC, Employee of: Samumed, LLC, Alyssa O'Green Shareholder of: Samumed, LLC, Employee of: Samumed, LLC, Carine Bossard Shareholder of: Samumed, LLC, Employee of: Samumed, LLC, Tim Seo Shareholder of: Samumed, LLC, Employee of: Samumed, LLC, Lisa Lamangan Shareholder of: Samumed, LLC, Employee of: Samumed, LLC, Maureen Ibanez Shareholder of: Samumed, LLC, Employee of: Samumed, LLC, Abdullah Ghias Shareholder of: Samumed, LLC, Employee of: Samumed, LLC, Carolyn Lai Shareholder of: Samumed, LLC, Employee of: Samumed, LLC, Long Do Shareholder of: Samumed, LLC, Employee of: Samumed, LLC, Shawn Cho Shareholder of: Samumed, LLC, Employee of: Samumed, LLC, Joseph Cahiwat Shareholder of: Samumed, LLC, Employee of: Samumed, LLC, Kevin Chiu Shareholder of: Samumed, LLC, Employee of: Samumed, LLC, Melinda Pedraza Shareholder of: Samumed, LLC, Employee of: Samumed, LLC, Yusuf Yazici Shareholder of: Samumed, LLC, Consultant for: Celgene Corporation, BMS, Genentech, Sanofi, Employee of: Samumed, LLC

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OP0073 ESTABLISHMENT OF HUMAN INDUCED PLURIPOTENT STEM CELL-LINES (IPSC) FOR IN VITRO MODELLING HAND OSTHEOARTHRITIS

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Background: Research results in the field of hand OA are currently limited due to the unavailability of tissue samples and lack of animal models replicating the features of the disease in humans. Cellular *in vitro* models are important tools to elucidate molecular mechanisms and pathways that are involved in hand OA. Specifically, induced pluripotent stem cells (iPSc) are considered ideal tools for this purpose since they allow the use of unlimited cells with chondrogenic differentiation potential. However, there are not studies published generating iPSc from patients with hand OA.

Objectives: To generate and characterize iPSc-lines from patients with radiographic hand OA and healthy donors, which can be used as cellular models of the disease, for studying the pathogenesis of the disease *in vitro* and for testing new drugs.

Methods: Patients with hand OA (non erosive hand OA with right thumb OA) and a healthy control were selected for the study. Using the explant culture technique, fibroblasts from skin biopsies of these patients were isolated. Transcriptional factors Oct4, Sox2, Klf4 and c-Myc were used for the reprogramming process; which was performed by using a non-integrating method, the Sendai virus. Cell lines obtained were morphologically, phenotypically and functionally characterized. To evaluate weather these iPSc lines could be used as cellular model of hand OA, presence of single nucleotide polymorphisms (SNPs) within the genes ALDH1A2 and SMAD3 were studied by Sanger sequencing, before and after reprogramming. Variants rs3204689 and rs12901499 respectively have been associated with severe OA of the hand (Styrkarsdottir *et al.*, 2014; Shu-Thao Gao *et al.*, 2018). Finally, chondrogenic differentiation capacity of the "healthy" and "ill" iPSc-lines was studied by means of histological techniques.

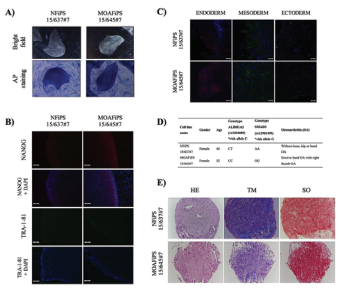

Figure 1. (A) Bright field images and images after alkaline phosphatase staining (AP staining) of iPSc colonies from each cell line. x4 magnification (B) Immunofluorescence images of pluripotency markers (NANOG and TRA-1-81) of representative clones from each cell line. Escale 100 µm. (C) Immunofluorescence images of endoderm, mesoderm and ectoderm markers in each one of the cell lines. Escale 100 µm. (D) Summarize of the results obtained after single nucleotide polymorphism analysis. (E) Haematoxilin-Eosin (HE), Masson's Tricromic (TM) and Safranin-O (SO) staining after chondrogenic differentiation of the iPSc lines. x10 magnification.

Results: Fibroblasts were isolated from one patient with radiographic hand OA and one healthy donor. Three weeks after reprogramming, embryonic stem cell-like colonies emerged in culture. These cells showed positivity for alkaline phosphatase activity (fig. 1A) and the pluripotency markers Tra1-81 and Nanog (fig. 1B). Molecular analyses showed high relative expression levels of the pluripotency-related genes OCT4, SOX2, NANOG and CRIPTO in the iPSc. These cells were also able to give rise to cells from the three germ layers (fig. 1C). Indeed, during mesodermal differentiation, spontaneously beating cardiomyocytes were seen in culture. Regarding SNPs studies, cells from the patient with hand OA were homozygous for the at-risk allele in both genes studied, both before and after reprogramming (fig. 1D). The "ill" iPSc-line (MOAFiPS 15/645#7) showed worse chondrogenic differentiation than the "healthy" iPSc-line (NFiPS 15/637#7), as shown by the micromasses collagen and proteoglycan content (fig. 1E).

Conclusion: The generation of one iPSc-line form patients with hand OA is reported for the first time. The presence of the at-risk alleles within the ALDH1A2 and SMAD3 genes were maintained after fibroblast reprogramming. The iPSC lines obtained shown differences in their chondrogenic differentiation capacity, showing their usefulness to model hand OA *in vitro*, and to deeper study the role of these genetic variants in the pathogenesis of hand OA.

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OP0074 TOFACITINIB PROMOTES FUNDAMENTAL PROCESSES OF BONE HEALING

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Background: Inflammatory diseases like rheumatoid arthritis (RA) and anti-inflammatory treatment of RA with glucocorticoids (GC) or NSAIDS negatively influence bone metabolism and fracture healing. Janus kinase (Jak) inhibition with tofacitinib has been demonstrated as a potent anti-inflammatory therapeutic agent in the treatment of RA but its impact on the fundamental processes of bone regeneration such as recruitment of human mesenchymal stromal cells (hMSCs) and chondrogenesis, osteogenesis and osteoclastogenesis is still controversial and in part elusive.

Objectives: Therefore, in this study, we aim to examine the effects of Tofacitinib on processes of bone healing under reduced oxygen availability mimicking the in vivo situation of the fracture gap.

Methods: To this end, we analyzed the influence of Tofacitinib on the (i) invasion of hMSCs towards TNFα using a trans-well assay, (ii) chondrogenic differentiation of hMSCs in a 3D-micro-mass culture under hypoxic conditions, (iii) osteogenic differentiation of hMSCs, (iv) M-CSF/RANKL-mediated differentiation of isolated monocytes towards osteoclasts and (v) hypoxia-mediated target gene expression in non-differentiated, osteogenic and chondrogenic differentiated hMSCs.

Results: We demonstrate that Tofacitinib dose-dependently promotes the recruitment of hMSCs under hypoxia but inhibits recruitment of hMSCs under normoxia. With regard to the chondrogenic differentiation of hMSCs, we observed that Tofacitinib did not inhibit survival and at therapeutic relevant doses of 10-100nM. Moreover, Tofacitinib dose-dependently enhances osteogenic differentiation of hMSCs and reduces osteoclast survival and differentiation under hypoxic conditions. We show that Tofacitinib does not affect chondrogenic HIF target gene expression but increases HIF target gene expression in human osteogenic differentiated MSCs.

Conclusion: We conclude from our data, that Tofacitinib may influence bone healing by promotion MSC recruitment into the hypoxic microenvironment of the fracture gap, does not interfere with the cartilaginous phase of the soft callus phase of fracture healing process. We assume that Tofacitinib may promote bone