Disclosure of Interests: Dornati Anang: None declared, Tamara H Ramwadhoeb: None declared, Janine Hahnlein: None declared, Bo van Kuik: None declared, Smits Noritje: None declared, Krijn P van Lienden: None declared, Mario Mas: None declared, Danielle Gerlag Employee of: Employee of UCB Pharma. UCB pharma was not involved in this study, Paul-Peter Tak Employee of: Employee of Candel therapeutics. Candel therapeutics was not involved in the study, Niek de Vries: None declared, Lisa van Baarsen: None declared. DOI: 10.1136/annrheumdis-2022-eular.289

Exploring the origin of inflammation in spondyloarthritis

A. Hierarchical cluster analysis of the top 100 most significant DEGs (FDR < 0.1) modulated by IL-17Ai separates pre- and post-treated groups. Normalized and scaled log2 gene expression levels are shown. B. Pathway enrichment analysis through DAVID. Enriched (p < 0.05) gene ontology terms from IL-17Ai-induced up- and down-regulated DEGs in peripheral SpA synovium are shown.

To assess if this response is tissue- and/or treatment-specific, we compared changes in gene expression by IL-17Ai in PsA synovium versus psoriatic skin, and in the PsA synovium after IL-17Ai versus IL-12p40/IL-23p40 blockade. While many inflammation-related GO terms and KEGG pathways were over-represented in both tissues and treatments, NFκB-, JAK-STAT-, and PI3K-Akt-signalling were enriched in DEGs in both skin and synovium after IL-17Ai, whereas JNK cascade, IL-17 signalling pathway and Th17 cell differentiation were over-represented in DEGs after IL-17Ai in the synovium specifically. Remarkably, IL-17Ai, but not IL-12p40/IL-23p40 blockade, modulated multiple bone-remodelling related pathways. Also, IL-17Ai modulated ossification and collagen catabolic process terms in PsA synovium and psoriatic skin in the opposite direction: these terms were over-represented in downregulated genes in synovium, but in upregulated genes in skin. Accordingly, genes upregulated after IL-17Ai were enriched in negative regulation of osteoblast differentiation in the synovium, but in positive regulation of osteoblast differentiation in the skin. These first in vivo human data provide molecular confirmation of previously reported animal data[3] that demonstrated down-modulation of disease-relevant immune and stromal pathways in the synovium in response to IL-17Ai.

REFERENCES:


Disclosure of Interests: Renée Fiechter: None declared, Leonieke van Mens: None declared, Ihsan Hammoura: None declared, Henriëtte de Jong: None declared, Desire Pots: None declared, Inka Fluri: None declared, Sander Tas: None declared, Dominique Baeten Employee of: Current employee of UCB Pharma, Mariëne G.H. van de Sande Consultant of: Novartis and AbbVie, Grant/research support from: Janssen, Novartis and Eli Lilly, Nataliya Yeremenko: None declared. DOI: 10.1136/annrheumdis-2022-eular.1516

OP001 MECHANICAL LOADING-INDUCED BHLHE40 PROMOTES INFLAMMATORY ARTHRITIS

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Background: Force induced microdamage to joint tissue is hypothesized to trigger inflammatory events in the joint leading to arthritis. Patients with inflammatory arthritis, such as rheumatoid arthritis (RA) and spondyloarthritis (SpA), are found to have inflammation in “mechanical hotspots” and mechanical loading in mouse models of these diseases is pro-arthritisogenic. To date, the molecular mechanism involved in converting a force to a biological signal that promotes arthritis is not known.

Objectives: This study aims to identify stretch induced genes in synovial fibroblasts, and the effect of these “mechano-sensitive” genes on arthritis.

Methods: Human synovial fibroblasts were stretched in vitro for 4hrs using the FlexCell system and analysed by microarray. Top stretch induced genes were measured in RA, SpA and healthy synovial tissue by qPCR. Patient synovium was further analysed by immunohistochemistry. Bhlhe40 deficient mice were subjected to collagen induced arthritis (CIA) and Kbxn serum transfer arthritis (STA). FACS was performed on ankle synovium. uCT was performed on whole ankles, with morphological changes scored by blinded readers, and calcaneus erosions by custom scripts in Fiji.

Results: 600 genes were found to be differentially expressed in stretched synovial fibroblasts (fold change > x1.5, adjusted p<0.05). 25% of these genes were found to be transcription factors, which included BHLHE40. BHLHE40 mRNA was elevated in the synovial tissue of RA/SpA vs healthy subjects (1.56 fold change), and BHLHE40 protein was widely detectable in synovial fibroblasts and macrophages (Figure 1). Bhlhe40 deficient mice were completely protected against CIA (incidence: 0% vs 40%, n=30 per group), but Bhlhe40 knockout did not block the generation of anti-collagen antibodies. Bhlhe40 deficient mice were partially protected against STA (peak clinical score at day 7: 5.2 vs 8.8, n=15 per group), with reduced synovial macrophage counts (CD11b+Ly6G-F4/80+) and neutrophil (CD11b+Ly6G+) frequency observed in the arthritic Bhlhe40 deficient mice compared to wildtype controls. Bhlhe40 had no impact on bone erosions with STA.
Figure 1. BHLHE40 is widely expressed in human synovium. Synovium obtained from total knee replacement. FFPE samples were stained for synovial macrophages (HLADR+) and fibroblasts (FAP+). Images acquired with the Zeiss LSM 780.

Conclusion: BHLHE40 was identified as a force-induced gene in synovial fibroblasts and was found to be upregulated in patients with inflammatory arthritis. Importantly, Bhlhe40 strongly promotes joint inflammation in murine models of arthritis and uncouples systemic autoimmunity from joint tissue inflammation. Thus, we have identified BHLHE40 as a novel regulator of mechanical load-associated inflammation.

REFERENCES:

Disclosure of Interests: None declared


THE SYNOVIIUM IN CHRONIC INFLAMMATORY JOINT DISEASES: COMPARISON OF CLINICAL, HISTOLOGICAL AND SCRNA-SEQ DATA BETWEEN RA, PSA, SPA AND UA

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Background: The synovium is the primary location of inflammation in various rheumatic diseases. However, specific differences of joint inflammation have not been explored on a single-cell level so far.

Objectives: To characterize the synovium of rheumatoid arthritis (RA), psoriatic arthritis (PsA), spondyloarthritis (SpA) and undifferentiated arthritis (UA) based on clinical and histological characteristics and single-cell RNA sequencing (scRNA-seq).

Methods: We performed ultrasound (US) guided synovial biopsy in patients fulfilling classification criteria: 10 RA, 4 PsA, 4 SpA and 3 UA. 3 osteoarthritis (OA) samples were obtained from surgery. Clinical data were collected at time of biopsy. Data analysis was performed using Cell Ranger, Seurat and Harmony R packages. scRNA-seq libraries were prepared with 10X Genomics on NovaSeq 6000. Gene expression of the synovium included Krenn score [1], synovial pathotype [2], vascularization [3] and presence of lymphoid follicles. OA histology was not available.

Results: We were able to compare the synovium of the most common chronic inflammatory joint diseases on various levels for the first time. The findings set the path for future diagnostic, prognostic, and therapeutic approaches in inflammatory joint diseases.

Disclosures of Interests: None declared


Table 1. Top 5 synovial fibroblast (SF) marker genes.

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Conclusion: We were able to compare the synovium of the most common chronic inflammatory joint diseases on various levels for the first time. The findings set the path for future diagnostic, prognostic, and therapeutic approaches in inflammatory joint diseases.

REFERENCES:
[2] Humby et al., JRD 2019
[3] Kennedy et al., AR 2010
[6] Park et al., JRD 2016

Figure

Disclosure of Interests: None declared