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Background: Recently a group of T-cell clones with characteristic T-cell receptor (TCR) motif was identified in peripheral blood and synovial fluid of HLA-B*27+ patients with ankylosing spondylitis (AS) [1-2] - a prototypic disease from a wider group of spondyloarthropathies (SpAs). Extraarticular manifestations of AS could involve skin, intestine or eye. Emerging data indicate linkage between intestinal and joint inflammation, including expression of gut-associated integrins on synovial T-cells [3-4]. However, clonal T-cell composition and presence of identical clones in different inflamed sites in SpAs remains poorly studied.

Objectives: To investigate clonal T-cell repertoire and presence of AS-related TCR motif in different sites of inflammation of patients with SpA.

Methods: Samples of synovial fluid (SF) were obtained from HLA-B*27+ and HLA-B*27- patients with ankylosing spondylitis (AS) and psoriatic arthritis (PsA), as well as gut biopsy samples from patients with AS and Crohn's disease (AS/CD) or ulcerative colitis (AS/UC), and conjunctival swabs from patients with uveitis (Uv) and with or without articular manifestations (Table 1). Also SF and gut biopsy samples were obtained from HLA-B*27+ patients with juvenile idiopathic arthritis (JIA). For one patient PsA patient paired samples of SF and gut biopsy were obtained.

Table 1. Detection of the AS-related motif TRBV9_CASS[V/A/L/P][G/A][L/T/V][F/Y]STDTQYF_TRBJ2-3 in bTCR repertoires of samples from different inflamed sites of patients with SpA

Tissue	Diagnosis	B27+	B27-	AS-related TCR motif+ among all samples from B27+ donors
Synovial fluid	AS PsA JIA	20	12	
Intestinal biopsy	AS/CD AS/UC JIA	4	3	3 / 4
Conjunctival swab	Uv	8	0	4 / 8

SF and gut samples were processed to isolate mononuclear cells, while conjunctival swabs were directly lysed in the lysis buffer. CD3+ β 7-intergrin+ cells were isolated from SF by fluorescence-activated cell sorting. Deep TCR repertoire profiling was carried out using UMI-based cDNA library preparation technology [1].

Results: Identical T-cell clonotypes were detected between paired SF and gut samples of the same patient with psoriatic arthritis and intestinal inflammation. The subpopulation of β 7-intergrin+ SF T-cells shared significantly more identical clonotypes with gut biopsy repertoire compared to the bulk SF T-cell repertoire. Clonotypes belonging to the AS-related TCR beta motif TRBV9_CASS[V/A/L/P][G/A][L/T/V][F/Y]STDTQYF_TRBJ2-3 were detected in all inflamed tissues tested: synovial fluid, intestinal biopsies and conjunctival swabs of SpA patients (Table 1). Importantly, we observed these clonotypes exclusively in samples from HLA-B*27+ donors (n=26), but not in HLA-B27- context (n=15) with comparable analysis depth, thus confirming strong HLA-B*27-restriction of the clonotypes. The AS-related clonotypes were detected in the subpopulation of β 7-intergrin+ SF T-cells from HLA-B*27+ patients with PsA.

Conclusion: For the first time we directly report the T-cell clonal sharing between synovial fluid and inflamed gut tissue of SpA patients. In sum our data suggests involvement of identical T-cell clones in inflammation in different anatomical sites in SpA.

REFERENCES:

- [1] Komech et al. *Rheumatology* (Oxford). 2018;57(6):1097-1104.
- [2] Faham et al. *Arthritis Rheumatol*. 2016;11(10):300-308.
- [3] Guggino et al. *Ann Rheum Dis*. Published Online First: 18 October 2019. doi:10.1136/annrheumdis-2019-216456.
- [4] Qaiyum et al. *Ann Rheum Dis*. 2019;78(11):1566-1575.

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OP0028

CD206+CD163+ PATHOGENIC MACROPHAGES ENRICHED IN RHEUMATOID ARTHRITIS SYNOVIAL TISSUE WITH DISTINCT TRANSCRIPTIONAL SIGNATURES

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Background: Synovial tissue macrophages are an exquisitely plastic pool of innate cells that play a key role in RA disease progression. However, the precise nature, diversity, and function of macrophage subsets within the inflamed joint remains unexplored.

Objectives: Therefore, the aims of this study are to phenotypically, transcriptionally and functionally characterise synovial tissue macrophages residing within the inflamed joint.

Methods: Rheumatoid Arthritis, Psoriatic Arthritis, Osteoarthritis and healthy control synovial-tissue biopsies and synovial-fluid mononuclear cells were analysed using the following panel (CD40, -CD45, -CD64, -CD68, -CD163, -CD206, -CD253, -CCR4, -CCR7, -CXCR1, -CXCR3). CD206+CD163+ and CD206-CD163- macrophages were sorted from RA synovial-tissue by FACSaria sorter; RNAseq and FLIM analysis, autologous T-cell co-culture and healthy fibroblast experiments performed. Cytokine expression was measured by MSD immunoassay.

Results: RA synovial tissue and fluid macrophages display markers typical of both M1 (CD40+CD253+) and M2 (CD206+CD163+) macrophages with a spectrum of macrophage activation states identified. Within this spectrum, significant enrichment of dominant CD206+CD163+ macrophage-subtype is present in synovial tissue versus fluid (p<0.05). CD206+CD163+ synovial tissue macrophages express significantly more CD40 than synovial fluid (p<0.0003), positively correlate with disease activity (r=0.6, p<0.01), with baseline levels predicting response to therapy (p<0.05). Moreover, CD206+CD163+CD40+ macrophages are enriched in RA synovial tissue compared to PsA and OA pathotypes (p<0.05). While the CD206+CD163+ subset is present in healthy synovial tissue, expression of CD40 is completely absent in healthy synovium (p<0.05) with dramatically decreased expression of CX3CR1 on RA macrophages. RNA-seq analysis indicates that CD206+CD163+ population is transcriptionally distinct from synovial tissue CD206-CD163-, synovial fluid CD206+CD163+, and RA monocyte-derived M1/M2 macrophages, with unique tissue-resident gene signatures. Moreover, differing metabolic demands between CD206+CD163+ and CD206-CD163- subsets was demonstrated by RNAseq and FLIM analysis. CD206+CD163+ macrophages enhance autologous T-cell responses, spontaneously secrete high levels of pro-inflammatory cytokines and activate healthy fibroblasts towards pro-inflammatory mechanisms thus further contributing to the local inflammatory response. Finally, inhibition of CD40 activity abrogates the expression of pro-inflammatory mediators (TNF α , IL-1 β , IL-6, IFN γ) and induces IL-10 expression in sorted CD206+CD163+ synovial tissue-macrophages suggesting a key role for CD40 in driving this pathogenic phenotype.

Conclusion: This data identifies for the first-time enrichment of a previously undescribed dysfunctional dominant and transcriptionally distinct macrophage subtype in RA synovial tissue. Taken together, this data provides a greater understanding of the critical role tissue-resident macrophages play in perpetuating inflammation in RA. Further investigation of the molecular patterns and cues that shape specific synovial macrophage subsets may provide opportunities to reinstate RA joint homeostasis.

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OP0029

NEUTROPHIL EXTRACELLULAR TRAPS IN GIANT CELL ARTERITIS INFLAMED TEMPORAL ARTERIES: LOCALIZATION, CO-EXPRESSION OF INFLAMMATORY CYTOKINES AND ASSOCIATION WITH DISEASE EXTENSION

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Background: Giant cell arteritis (GCA) represents the most prevalent form of systemic vasculitis in elderly, characterized by a remarkable heterogeneity in terms of clinical and histological phenotype, the pathogenetic mechanisms and treatment selection (1). An interplay between cells of both innate and adaptive immunity, appear to govern the pathophysiological mechanisms (2). Among the different cellular populations, neutrophils may hold a central role in GCA pathogenesis, since they are present in abundance in tissue injury (3,4). Most importantly, they are a source of neutrophil extracellular traps (NETs) that may deliver immunocompetent substances, necessary for the perpetuation of the inflammatory response (5). The detection and function of NETs have not been studied in GCA. **Objectives:** To explore the presence and clinical significance of NETs in temporal artery biopsies (TABs) of patients with GCA.

Methods: Ten patients with GCA [5 with limited cranial vasculitis (CV) and 5 with associated generalized large vessel vasculitis (LVV), as defined by ¹⁸F-fluorodeoxyglucose (FDG) positron-emission tomography with computed tomography (PET/CT)] and 8 patients with polymyalgia rheumatica (PMR) were studied. GCA and PMR patients fulfilled the 1990 ACR and 2012 EULAR/ACR provisional classification criteria, respectively. The presence, location, quantitation and decoration of NETs with IL-6, IL-1 β , and IL-17A were assessed in TABs at the time of disease diagnosis by tissue immunofluorescence and confocal microscopy. Quantification of NETs in tissue sections was performed using the Imaris v.9.3 software that counts the total measure volume instead of only the projection area (6). Serum levels of IL-6 and IL-17A around the time of tissue biopsy were also evaluated in all patients.

Results: All temporal artery biopsies from GCA patients had NETs located mainly in the adventitia, adjacent to the vasa vasorum, whereas TABs from PMR patients had no NET structures. LVV was associated with a higher NETs to total tissue volume ratio, compared to CV-GCA [$p=0.0317$]. NETs decorated with IL-6 were present in TABs of all LVV and 3 of 5 CV-GCA patients, while IL-17A positive NETs were observed in all GCA patients. IL-1 β -positive NETs were not detected in any GCA patient. No relation was found between serum IL-6 and IL17A levels and NETs containing IL-6 and/or IL-17A.

Conclusion: NETs bearing IL-6 and IL-17A cytokines are present in inflamed GCA-TABs. IL-6 positive NETs are associated with the LVV phenotype and might be useful as a tissue biomarker for disease severity and extent.

REFERENCES:

- [1] K. S. M. van der Geest et al. *Arthritis Rheumatol*, 2018;70:1366-76.
- [2] C. Salvarani et al. *Lancet*, 2008;372:234-45.
- [3] D. Chatelain et al. *Ann Rheum Dis*, 2009;68:84-8.
- [4] S. Nadkarni et al. *Circ Res*, 2014;114:242-8.
- [5] V. Mutua et al. *Clin Rev Allergy Immunol*, 2020.
- [6] S. V. Costes et al. *Biophys J*, 2004;86:3993-4003.

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Of mice and man: insights into pathophysiology of rheumatoid arthritis and spondyloarthritis

OP0030

TREATMENT OF NON-BIOLOGIC-DMARD-IR PSA PATIENTS WITH UPADACITINIB OR ADALIMUMAB RESULTS IN THE MODULATION OF DISTINCT FUNCTIONAL PATHWAYS: PROTEOMICS ANALYSIS OF THE SELECT-PSA 1 PHASE 3 STUDY

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Background: Treatment of non-biologic-DMARD-IR (DMARD-IR) PsA patients with upadacitinib (UPA) at 15 mg QD, an oral JAK1 selective inhibitor, resulted in improvement in musculoskeletal symptoms, psoriasis, physical function, fatigue, quality of life, and inhibited radiographic progression; improvements were

observed as early as Week 2 (ACR20 and ACR50). UPA 15 mg QD was non-inferior to adalimumab (ADA) 40 mg EOW for ACR20¹

Objectives: To determine the relative biological pathway modulation of UPA compared with ADA in patients with PsA via the evaluation of a pre-defined set of plasma proteins associated with inflammation.

Methods: Patients from the SELECT-PSA1 study (DMARD-IR PsA patients) were randomly selected (PBO, n=100; UPA 15 mg QD, n=100; ADA 40 mg EOW, n = 100). The levels of 92 inflammation related protein biomarkers (pBM) were analyzed using a multiplexed Proximity Extension Assay platform in plasma samples collected at baseline, week 2, and 12; change from baseline in protein levels were expressed as Log₂ Fold Change; a Repeated Measure Mixed Linear Model identified proteins modulated by UPA and ADA compared to Baseline. Functional pathway prediction was performed in silico with a commercially distributed software where 52 significantly modulated pBM (mean ILog₂ FCI ≥ 0.1 AND FDR ≤ 0.05) were selected; results were summarized based on 3 core biological groups: 1) adaptive immune system, 2) innate immune system, and 3) non-immune connective and vascular systems.

Results: At the single pBM-level, at the week 2 and 12 time points, treatment with UPA 15 mg QD resulted in distinct down modulation of T cell-associated (CD5, CD8a, IL15Ra, SLAMF1, TRANCE) and myeloid cell-associated pBM (CSF-1, CCL7, CCL13) that was not observed in the ADA treated group. Reciprocally, treatment with ADA 40 mg EOW resulted in a specific down modulation of a subset of neutrophil associated pBM (CCL3, CCL4, and S100A12). Both treatments resulted in the down modulation of IFN-, IL6-, and TNF-related pBM (CXCL9, CXCL10, CXCL11, IL6, TNFRSF19, and TNFSF14) suggesting a common node of activity related to these pivotal cytokine-signaling pathways.

Functional pathway prediction based on the pBM data revealed that treatment with UPA is preferentially associated with the inhibition of T cells, but also NK cells and lymphocytes, compared to the predicted effects of treatment with ADA. Treatment with UPA also preferentially inhibited pathways related to bone damage and angiogenesis, as compared to the predicted effect of treatment with ADA. Finally, both treatments were predicted to inhibit multiple pathways associated with the activity of myeloid cells and phagocytes.

Conclusion: Consistent with previous observations in RA², UPA is predicted to inhibit multiple functional pathways associated the pathobiology of PsA belonging to the general categories of adaptive and innate immunity but also non-immune vascular and connective tissue biology. In contrast, treatment with ADA appears to affect more specifically functional pathways associated with the innate immune system.

REFERENCES:

- [1] McInnes, I. et al. *Annals of the Rheumatic Diseases* 79, 16-17 (2020).
- [2] Sornasse, T., Song, I.H., Radstake, T. & McInnes, I. *Annals of the Rheumatic Diseases* 79, 581-582 (2020).

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