P063/O11 INHIBITION OF ARGINASE-1 EXPRESSION BY THE TRANSCRIPTION FACTOR FRA-1 IN MACROPHAGES EXACERBATES RHEUMATOID ARTHRITIS INFLAMMATION

¹N Hannemann*, ¹S Cao, ¹A Schnelzer, ²J Jordan, ³M Eberhardt, ⁴U Schleicher, ⁵S Uebe, ⁶A Ekeci, ¹J Rech, ²T Bäuerle, ⁴C Bogdan, ³J Vera, ¹G Schett, ¹A Bozec. ¹Department of Internal Medicine 3; ²Institute of Radiology, Preclinical Imaging Platform Erlangen (PIPE); ³Department of Dermatology; ⁴Institute of Microbiology – Clinical Microbiology; ⁵Institute of Human Genetics Universitätsklinikum Erlangen; ⁶Institute of Human Genetics, erlangen, Germany

10.1136/annrheumdis-2018-EWRR2019.53

Career situation of first and presenting author Student for a master or a PhD.

Introduction The activator protein (AP)-1 transcription factor family, especially its subfamily of FOS proteins (cFos, FosB, Fra-1 and Fra-2) are associated to the regulatory network of macrophage responses. Moreover, it is well known that macrophages are central player during rheumatoid arthritis (RA).

Objectives This study aims to delineate the role of Fra-1 in macrophages during the acute destructive inflammatory phase of RA.

Methods Therefore, we applied the serum-induced arthritis (K/BxN) model to Fra-1 deficient mice controlled by the Mx1 promoter (Fra-1^{ΔMx}) or the LysM promoter (Fra-1^{ΔLysM}). Moreover, we performed *in vitro* analyses of macrophage polarization in wildtype and Fra-1 deficient macrophages, as well as micro-arrays and ChIP sequencing analyses to delineate Fra-1 targets in activated macrophages. We completed our analysis by studying Fra-1 expression in RA patients' synovium.

Results Fra-1 mutant mice had decreased arthritis severity compared to their littermate wildtype mice. The alleviated arthritis was accompanied to increased arginase-1 (Arg1) expression and activity in the joints, suggesting that its anti-inflammatory features milder RA inflammation. Sorting of immune cell populations revealed macrophages as the major source of Arg1, which was increased in Fra-1 mutant mice. Mechanistically, Fra-1 transcriptionally inhibited Arg1 expression in macrophages. Moreover, inhibition of Arginase in Fra-1 mutant mice restored a full blunt inflammatory RA response and the supplementation of mice with l-arginine, leading to increased arginase activity in the joint, is sufficient to milder arthritis. Synovium histological sections from RA patients showed a correlation between Arg1, Fra-1 and the DAS28 score, confirming that increased Arg1 activity is of benefit also for human inflammatory joint disease.

Conclusions Our data show for the first time that Fra-1 is a pivot between pro- and anti-inflammatory macrophage. By inhibiting Arg1 activity, Fra-1 exacerbates RA inflammation and joint destruction.

Disclosure of Interest None declared.

P065 REGULATION OF JOINT DESTRUCTION BY ACTIVIN A IN RHEUMATOID ARTHRITIS

V Kracke*, M Fennen, J Intemann, E Werbenko, D de Gorter, P Paruzel, T Pap, B Dankbar. Institute of Musculoskeletal Medicine, Münster, Germany

10.1136/annrheumdis-2018-EWRR2019.54

Career situation of first and presenting author Student for a master or a PhD.

Introduction Activins and inhibins belong to the transforming growth factor β family. Activins are disulphide-linked homo- or heterodimers consisting of two inhibin β chains (β A, β B) that are expressed in many cell types. However, activin A (β A β A) is the only activin that is expressed in bone and cartilage. Moreover, activin A has been demonstrated not only to stimulate receptor activator of NF- κ B ligand (RANKL)-induced osteoclast (OC) differentiation but also to inhibit osteoblast differentiation.

Objectives Here we investigate the impact of activin A on joint destruction in rheumatoid arthritis.

Methods Synovial tissue samples from rheumatoid arthritis (RA) and osteoarthritis (OA) patients were analysed by immunohistochemical staining. For *in vitro* experiments, synovial fibroblasts (FLS) were isolated from hind paws of WT mice. Effects of cytokines on the secretion of activin A by mouse FLS were evaluated by ELISA. Bone marrow-derived macrophages (BMM) were isolated from femurs and tibias of WT mice and differentiated into osteoclasts in the presence of macrophage colony-stimulating factor (M-CSF) and RANKL with or without activin A. OC differentiation was characterised by TRAP staining. Resorption activity was determined by quantification of osteoclast-mediated pit formation on a calcium phosphate-coated plate. Furthermore, osteoclast-specific gene expression as well as the activation of SMAD2 in BMMs, OCs and FLS were analysed by immunoblotting. The interaction of phospho-SMAD2 with NFATc1 was evaluated by co-immunoprecipitation using Dynabeads.

Results We demonstrate that activin A is highly abundant in the synovium of RA but not of OA patients. *In vitro*, activin A secretion by FLS was strongly enhanced by pro-inflammatory cytokines. Furthermore, activin A strongly enhanced the RANKL-mediated differentiation of BMMs into mature OCs, reflected by a significantly increased OC number and OC size. Moreover, concomitant administration of activin A led to a significant increase of the total resorption area as well as resorption area per pit, indicating an increased activity of individual OCs. Furthermore, activin A enhanced the RANKL-induced expression OC differentiation markers, but alone was not able to induce OC differentiation. Analyses of signaling pathways revealed that activin A induce the activation of SMAD2 in BMMs and OCs. Finally, upon co-stimulation with RANKL, activin A resulted in an increased interaction between activated SMAD2 and NFATc1.

Conclusions The data strongly suggest that increased expression of activin A in the arthritic joint is associated with enhanced osteoclast formation, promoting joint destruction in rheumatoid arthritis.

Disclosure of Interest None declared.

P066 MIR-342-3P PROMOTES CELL SURVIVAL AND MOTILITY OF OSTEOCLAST PRECURSORS

^{1,2,3}C Lozano*, ^{1,3}V Estivals, ^{1,3}I Duroux-Richard, ^{1,2,3}F Apparailly. ¹U1183-IRMB, INSERM; ²CHU Montpellier; ³University Montpellier, Montpellier, France

10.1136/annrheumdis-2018-EWRR2019.55

Career situation of first and presenting author Young investigator.

Introduction Chronic inflammatory joint disorders are associated with bone destruction by osteoclasts (OC), which derive from myeloid precursors. Recent findings reveal that OC are