4 weeks): (I) 8 week old mice with established inflammatory, erosive arthritis and (II) 12 week old hTNFtg mice with severe inflammatory, erosive arthritis (n=8–10 animals). Clinical signs of arthritis were weekly assessed. Knee and ankle joints were used for μ CT (Scanco μ CT35) and subsequent histology. Toluidine blue staining indicated proteoglycan contents, tartrate-resistant acid phosphatase staining identified osteoclasts. Immunohistochemical stainings were performed for collagen type II (col II) and osterix. Expression of chondrogenic markers was analysed from RNA paw extracts.

Results TNF blockade significantly improved clinical signs in both treatment cohorts. Compared to eroded bone surfaces at week 8 (before treatment), TNF blockade led to improved bone architecture with regular, smooth bone surfaces in CT scans from knee and ankle. Histologically, treatment allowed complete resolution of synovitis, osteoclast activity and subchondral bone erosions. Despite sites of intact bone tissue, regenerative processes of cortical and subchondral bone were found. Erosions of bone and calcified cartilage were filled with col II positive cartilaginous or fibro-cartilaginous tissue. Hyperproliferative chondrocytes and endochondral ossifications were found in knees. Consistently, chondrogenic markers like Sox9 transcription factor and col II were markedly increased, accompanied by decreased expression of cartilage degrading enzymes (MMP3, MMP13) upon treatment. Of note, menisci and patella tendon showed massive hyperproliferative chondrocytes, col II and osterix staining.

Anti-TNF treatment at later stages with severe bone destruction (at week 12) could not completely resolve synovitis and restore structural damage. Pannus tissue transformed into chondrocyte rich, fibrocartilaginous tissue. Callus and ossifications were formed in knees.

Conclusions Therapeutic TNF blockade of inflammation-mediated joint damage enables the initiation of directed but also undirected regenerative processes. Remission of synovitis seems to be essential to promote bone and calcified cartilage repair.

Disclosure of Interest None declared.

P115/O19 IMMUNOMODULATORY ACTIVITY OF ADIPOSE-DERIVED MESENCHYMAL STEM CELLS OF ANKYLOSING SPONDYLITIS PATIENTS

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Introduction Ankylosing spondylitis (AS) is a chronic autoimmune disease characterized by inflammation and pathological new bone formation at axial joints with resulting spinal segments fusion. The immune abnormalities observed in AS patients concern mainly T lymphocytes and manifest by increased numbers of circulating Th17 cells and defects of regulatory T (Treg) cells. Adipose mesenchymal stromal/stem cells (ASCs) exert

immunosuppressive effects on different immune cells, including T lymphocytes, and may be promising option for successful therapy of AS.

Objectives In this study we have focused on immunoregulatory activity of ASCs from AS patients (AS/ASCs) toward healthy donor peripheral blood mononuclear cells (PBMCs) and T lymphocytes with special regard to Th17 and Treg cell subsets.

Methods AS/ASCs of 15 AS patients and commercially available ASC lines from healthy donors (hASC) were used. ASCs were co-cultured with either anti-CD3/CD28-stimulated CD4⁺ T lymphocytes or mitogen-stimulated PBMCs of allogeneic healthy volunteers. Expression of CD25 and FoxP3 transcription factor in T cells and these cells proliferation were evaluated by flow cytometry. Concentrations of cytokines associated with Th1 (IFN γ), Th17 (IL-17AF) or Treg (IL-10, TGF β) cells as well as of factors related to immunomodulatory function of ASCs (PGE₂, kynurenines) were measured by ELISAs in co-culture supernatants.

Results In the co-cultures of ASCs with PBMCs both hASCs and AS/ASCs exerted similar inhibitory effect on the proliferation response of T CD4⁺ and CD8⁺ cells. Similar results were obtained in co-cultures of ASCs with purified T CD4⁺ cells. In the co-cultures of hASCs and AS/ASCs with PBMCs as well as with purified T CD4⁺ cells the significant increases of kynurenines and PGE₂ production were observed. In co-cultures of ASCs and PBMCs there was a marked decrease of IFN γ and significant increase of IL-17AF. Concomitantly, Treg proportion in these co-cultures was elevated. In contrary, in the co-cultures of activated T CD4⁺ cells with ASCs there was some increase of IFN γ and IL-17AF whereas Treg proportion was downregulated.

Conclusions AS/ASCs and hASCs exert comparable effects on T cells. Since different effects were observed in the co-cultures of ASCs with PBMCs than with CD4⁺ cells, it seems that ASCs activity toward Th17 and Treg cells is dependent on the presence of accessory cells.

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P116 BASIC CHARACTERISTICS OF ADIPOSE-DERIVED MESENCHYMAL STEM CELLS OF ANKYLOSING SPONDYLITIS PATIENTS

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Career situation of first and presenting author Post-doctoral fellow.

Introduction Application of mesenchymal stem/stromal cells (MSCs), endowed with immunosuppressive and regenerative properties, may be promising option for successful therapy of ankylosing spondylitis (AS) patients, characterised by inflammation and pathological bone remodelling. Adipose tissue is an