Results: A total of 27 studies included 4,678 JIA cases and 7,634 control subjects. There were 23 studies on Caucasians, 3 on Asians, and 1 on Africans. Seven studies were on IL-1 α -899 C/T, 5 were on IL-1 β -511 C/T, 6 were on IL-6-174 G/C, 19 were on TNF- α -308 G/A, and 14 were on TNF- α -238 G/A polymorphisms. Meta-analysis of IL-1 α -899 C/T, IL-1 β -511 C/T, IL-6-174 G/C, and TNF- α -308 G/A and -238 G/A polymorphisms revealed no significant associations between JIA and minor alleles (OR 0.99, 95% CI 0.87–1.12, P = 0.84; OR 1.10, 95% CI 0.97–1.25, P = 0.15; OR 1.04, 95% CI 0.91–1.19, P = 0.57; OR 1.09, 95% CI 0.87–1.37, P = 0.46; OR 0.85, 95% CI 0.63–1.13, P = 0.26). Subgroup analysis was conducted on the relationship between JIA and proinflammatory cytokines based on ethnicity. In Caucasians, there was no significant association between IL-1 α -899 C/T, IL-6-174 G/C, and TNF- α -308 G/A and -238 G/A polymorphisms and JIA; however, there were significant relationships between JIA and recessive (TT vs. CT + CC) and additive (TT vs. CC) models of IL-1 β -511 C/T polymorphisms (OR 1.48, 95% CI 1.09–2.00, P = 0.01; OR 1.46, 95% CI 1.05–2.03, P = 0.02).

Conclusion: IL-1 α -899 C/T, IL-1 β -511 C/T, IL-6-174 G/C, and TNF- α -308 G/A and -238 G/A polymorphisms were not significantly associated with overall JIA susceptibility in all ethnicities. However, differences according to ethnicity were observed, and the TT genotype of IL-1 β -511 C/T was associated with a high prevalence of JIA in Caucasians.

Figure 1. OR and 95% CI of individual studies and pooled data for the association between minor alleles of TNF- α -308 G/A and JIA in all study subjects.

Abstract THU0037 – Figure 1

Figure 2. OR and 95% CI of individual studies and pooled data for the association between the IL-1 β -511 C/T polymorphisms and JIA in Caucasian subjects. (A) recessive and (B) additive models.

Abstract THU0037 – Figure 2

REFERENCES:

- Cimaz R, Cazalis MA, Reynaud C, Gerloni V, Zulian F, Biggioggero M, Martini G, Pontikaki I, Fantini F, Mouglin B, Miossec P (2007) IL1 and TNF gene polymorphisms in patients with juvenile idiopathic arthritis treated with TNF inhibitors. *Ann Rheum Dis* 66:900-904.
- Donn RP, Barrett JH, Farhan A, Stopford A, Pepper L, Shelley E, Davies N, Ollier WER, Thomson W (2001) Cytokine gene polymorphisms and susceptibility to juvenile idiopathic arthritis. *British Paediatric Rheumatology Study Group. Arthritis Rheum* 44:802-810.
- McDowell TL, Symons JA, Ploski R, Førre Ø, Duff GW (1995) A genetic association between juvenile rheumatoid arthritis and a novel interleukin-1 α polymorphism. *Arthritis Rheumatism* 38:221-228.

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THU0038

INDIVIDUAL FUNCTIONS OF THE HISTONE-ACETYLTRANSFERASES CBP AND P300 IN REGULATING THE INFLAMMATORY RESPONSE OF RHEUMATOID ARTHRITIS SYNOVIAL FIBROBLASTS

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Background: The two coactivators of transcription cAMP-response element binding protein (CREB) binding protein (CBP) and p300 are close homologues. They are widely accepted as redundant proteins and unique functions have not been investigated in depth. Both proteins contain a histone acetyltransferase (HAT) activity for writing of cell type specific activating H3K27 histone acetylation marks, and a bromodomain, which functions as a reader of acetylated lysine residues on histone tails. Inhibitors targeting the bromodomains are in drug development for inflammatory and malignant diseases.

Objectives: To analyze individual functions of CBP and p300 in regulating the inflammatory response of rheumatoid arthritis (RA) synovial fibroblasts (SF).

Methods: SF were isolated from knee, shoulder and hand joints of RA patients undergoing joint replacement surgery. The expression of CBP and p300 was silenced by transfection of antisense LNA gapmeRs (12.5 nM). 24h after transfection, cells were stimulated with TNF- α (10 ng/ml, 24h). The levels of H3K27ac in SF were analyzed by Western blotting (n=7). Transcriptomes were determined by RNA-seq (Illumina NovaSeq 6000, n=6). Pathway enrichment analysis of RNAseq data was performed using DAVID bioinformatic resources (fold change > 1.5, FDR < 0.05, top 3000 genes included). Changes in the mRNA expression of potential target genes were confirmed by quantitative Real-time PCR (n=12-14).

Results: Silencing of p300 reduced the levels of H3K27ac by 30% in unstimulated SF, and by 61.4% ($p < 0.05$) in presence of TNF- α , whereas silencing of CBP reduced H3K27ac by 43.5% only in presence of TNF- α . In line with the changes in the H3K27ac, silencing of p300 affected the expression of 6026 and 5138 genes in unstimulated and stimulated SF, respectively. In contrast, only 285 and 1911 genes were affected by CBP silencing in unstimulated and stimulated SF, respectively. 184 genes were affected by both, CBP and p300, in unstimulated SF, with 9.2% of genes being regulated in opposite directions. 13.5% of overlapping genes affected by CBP and p300 were regulated in opposite directions in TNF- α -stimulated SF. Principal component (PC) analysis of RNAseq data separated TNF- α -stimulated from unstimulated SF (PC1) and p300 gapmeR-transfected SF (PC2) from controls and CBP gapmeR-transfected SF, which clustered together. The top pathways regulated by CBP were 'cell cycle' and 'focal adhesion' in unstimulated cells and 'DNA replication' and 'cell cycle' after stimulation with TNF- α . Top pathways regulated by p300 in presence and absence of TNF- α were 'proteasome', 'spliceosome' and 'focal adhesion'. The expression of 16 chemokines and cytokines was changed in RNAseq data (fold change > 1.5, $p < 0.05$) by either silencing of CBP or p300. Whereas silencing of CBP reduced the expression of all of them, silencing of p300 had pro- and anti-inflammatory effects. We further confirmed expression changes in cytokine and chemokine expression by Real-time PCR. Silencing of CBP reduced the expression of IL6, CCL2 ($p < 0.01$), CXCL31 ($p < 0.05$), and CXCL10. Silencing of p300 reduced the expression of CCL2 and CXCL31 ($p < 0.001$) but increased the expression of IL8 ($p < 0.001$) and CXCL2 ($p < 0.05$).

Conclusion: Our results suggest that p300 is the major writer for H3K27ac marks in SF. We have identified overlapping and distinct functions for CBP and p300 in SF. CBP inhibition has anti-inflammatory effects. In contrast, p300 inhibition has pro- and anti-inflammatory functions.

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In addition, he had/had consultancy relationship within the last 3 years with A. Menarini, Amgen, Abbvie, GSK, Mepha, MSD, Pfizer and UCB in the field of arthritides and related disorders, Caroline Ospelt: None declared, Kerstin Klein: None declared

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THU0039 THE RELATION BETWEEN THE INFLAMMATORY STATUS OF HUMAN END STAGE OSTEOARTHRITIC SYNOVIUM AND LEVELS OF LOW DENSITY LIPOPROTEIN

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Background: High systemic levels of low density lipoprotein (LDL) is a shared metabolic risk factor for osteoarthritis (OA) and cardiovascular disease (CVD), which may point to common biochemical pathways. One of the key events in atherosclerosis is formation and uptake of oxidized LDL (oxLDL) as a consequence of macrophage-mediated inflammation. Studies performed in our lab have previously shown that in mice, high LDL levels enhance synovial activation and ectopic bone formation during development of collagenase-induced OA. Furthermore, injection of oxLDL in naïve macrophage depleted joints led to infiltration of myeloid progenitor cells. Although these results strongly indicate towards (ox)LDL-mediated events that enhance OA severity, these processes have not yet been shown to occur in human osteoarthritic synovium.

Objectives: In the present study, we investigated whether the inflammatory status of OA synovium is associated to systemic and local levels of (ox)LDL.

Methods: Blood was collected from patients planned to undergo total knee replacement and LDL serum levels were determined (n=16). After surgery, synovial explants from the same patients (2-8 per patient) were collected and incubated at 37°C for 24 hours in presence of LDL (50 µg ApoB/mL), oxLDL (50 µg ApoB/mL) or control medium. Levels of IL-1β, TNF-α, IL-10 and MCP-1 secreted in the conditioned medium were measured using Luminex. Levels of S100A8/A9 were measured using ELISA. Concentrations of secreted factors were normalized for tissue sample weight. Synovial explants were further processed for histological analysis. Immunohistochemical staining of ApoB was performed to assess presence and uptake of (ox)LDL in the synovium. Hematoxylin & eosin-stained sections were used to arbitrarily score cellular infiltration and synovial hyperplasia.

Results: Around 50% of the patients in our cohort showed synovial activation exemplified by high secretion levels of IL-1β, TNF-α and IL-10. Cellular infiltration and synovial hyperplasia was observed in the majority of synovial tissue samples, their grades did however not correlate with cytokine secretion. Secretion levels of IL-1β positively correlated with serum levels of LDL and total cholesterol and did not correlate with BMI, suggesting a link that runs via systemic metabolic processes rather than increased weight-mediated joint loading. Considering that around 50% of OA patients suffers from synovitis characterized by increased macrophage presence and activity, high LDL in these patients may trigger oxLDL formation and uptake in the synovium. Staining of LDL-associated protein ApoB in histological sections of synovia showed distinct speck-like staining within macrophages and endothelial cells especially, suggesting oxLDL accumulation in these cells. Unexpectedly, presence of high concentrations of LDL or oxLDL during incubation of synovial explants did not significantly affect secretion of IL-1β, TNF-α, IL-10 and MCP-1, whereas presence of LDL but not oxLDL negatively affected secretion of S100A8/A9.

Conclusion: IL-1β secretion by end stage OA synovium is associated to systemic levels of LDL. Intervention with high concentrations of LDL or oxLDL for 24-hours after synovium extraction did however not alter cytokine secretion, suggesting that LDL and synovitis are linked through more long-term local interactions, or alternatively via systemic inflammatory factors.

Disclosure of Interests:

None declared

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THU0040 USING A NOVEL BEAD-BASED IMMUNOASSAY FOR SIMULTANEOUS DETECTION OF AUTOANTIBODIES AGAINST SERUM AMYLOID A1 AND ALPHA1 ACID GLYCOPROTEIN

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Background: Naturally occurring autoantibodies (Abs) against acute phase proteins (APPs), such as anti-serum amyloid A1 (SAA1) Abs have already been identified in sera of healthy individuals, as well as in patients with autoimmune diseases (1). Currently however, no method exists for simultaneous detection of multiple Abs against APPs.

Objectives: To develop a bead-based duplex immunoassay for simultaneous detection of IgG anti-SAA1 and anti-alpha 1 acid glycoprotein (AGP) Abs and quantify their levels in sera of healthy blood donors (HBD), and patients with systemic autoimmune diseases, as well as in intravenous immunoglobulin preparation (IVIg).

Methods: Fluorescently labeled MagPlex microspheres (Luminex Corp) were used to covalently bind SAA1 and AGP. Sera samples (diluted 1:20) were added to a 96-well microtiter plate and incubated with SAA1- and AGP-coupled microspheres for 2h. Subsequently, PE-conjugated anti-human IgG Abs were added to each well, incubated for 30 min, resuspended in PBS-1%BSA and analysed using the MagPix system (BioMedica, GmbH).

Abstract THU0040 – Figure 1. Levels of antibodies against SAA1 (A) and AGP (B) in HBD as compared to patients with systemic autoimmune diseases. Shown is median and IQR. 5th and 95th percentile of MFI values in HBD are indicated with dotted lines; *p<0.05; **p<0.01. SAA1, serum amyloid A1; AGP, alpha 1 acid glycoprotein; MFI, median fluorescence intensity; HBD, healthy blood donors; GCA, patients with giant cell arteritis; IgAV, immunoglobulin A vasculitis; SLE, systemic lupus erythematosus; SSc, systemic sclerosis; APS, antiphospholipid syndrome; ERA, early rheumatoid arthritis.

Abstract THU0040 – Figure 2. Presence of antibodies against SAA1 and AGP in IVIg. IVIg, intravenous immunoglobulin G; SAA1, serum amyloid A1; AGP, alpha 1 acid glycoprotein; MFI, median fluorescence intensity.

Sera samples were collected from HBD (n=55), patients with giant cell arteritis (GCA; n=30), immunoglobulin A vasculitis (IgAV; n=30), systemic lupus erythematosus (SLE; n=20), systemic sclerosis (SSc; n=27), antiphospholipid syndrome (APS; n=19) and early rheumatoid arthritis (ERA; n=20). Ab levels were determined also in IVIg (Octapharma AG).

Results: We observed the presence of both, anti-SAA1, and anti-AGP Abs in sera of HBD, as well as in patients with GCA, IgAV, SLE, SSc, APS and ERA.

GCA patients had significantly higher anti-SAA1 levels (median (IQR) MFI was 2163 (1113- 3048)) as compared to SSc (816 (480-1462); p<0.01) and ERA (886 (722-1596); p<0.05) patients. Levels of anti-AGP Abs were also significantly higher in sera of GCA patients (495 (298-872)) as compared to ERA (213 (130-338); p<0.01) and APS (264 (154-399); p<0.05) patients. No difference in anti-SAA1 or anti-AGP levels was observed between HBD and the tested groups of patients (Figure 1).