A1.9 **PREDICTION OF TREATMENT RESPONSE BY ACPA LEVELS AMONG PATIENTS WITH RHEUMATOID ARTHRITIS**

Raquel Hernández, Paz Gonzalez, Julia Uceda, Jose Luis Marenco de la Fuente. *Valme University Hospital, Seville*

Background The relationship between anticitrullinated protein antibodies (ACPA) and the clinical outcome of rheumatoid arthritis (RA) under treatment has been evaluated in clinical trials with conflicting results. Thus, an analysis of the BeSt study indicated that response to treatment was similar in ACPA-positive and ACPA-negative patients (van den Broek *et al*, Ann Reum Dis 2012; 71: 245–248). However, ACPA-positive patients showed more frequently radiological damage progression. On the contrary, in the IMPROVED study, ACPA-negative patients achieved less remissions on treatment than ACPA-positive patients (Wevers-de Boer *et al*, Ann Reum Dis 2012). Because of these, we evaluated the clinical response of patients with RA receiving treatment according to ACPA titers.

Patients and Methods This was a retrospective longitudinal observational study. We included all patients seen at our Unit who fulfilled the following criteria: 1) Diagnosis of RA by a rheumatologist meeting the 1987 criteria for RA; 2) Available determinations of ACPA; 3) Treatment for AR (whether or not biological therapy) with a minimum follow up of 6 months. The outcome variable was clinical regression defined as reaching DAS28 ≤ 2.6. Predictors of the outcome variable were evaluated using logistic regression models adjusted by gender and age.

Results 71 patients were included, 79% of them women. Clinical regression was observed in 42 (59%) patients during the first 12 months of follow-up. Baseline median (IQR) ACPA levels were 363 (27.7–500) for individuals without clinical regression and 91.7 (7–458) for those with regression ($p = 0.045$). 19 (66%) patients without regression versus 15 (36%) with regression showed ACPA levels ≥200 ($p = 0.013$). 6 (20%) patients without clinical response versus 18 (42%) individuals with response showed negative ACPA titers ($p = 0.050$). Factors independently associated with clinical regression were: Recent onset RA [adjusted odds ratio (AOR) 5; 95% confidence interval (95%CI) 1.01–25; $p = 0.049$], baseline ACPA levels ≥200 [AOR 0.13; 95%CI 0.03–0.5; $p = 0.004$], baseline CRP levels [per unit increase; AOR 0.94; 95%CI 0.91–0.99; $p = 0.024$], baseline DAS28 [per unit increase; AOR 0.47; 95%CI 0.25–0.89; $p = 0.021$].

Conclusions ACPA levels can predict clinical regression of patients with RA receiving treatment in real life conditions. Individuals with high ACPA levels may benefit from a more aggressive treatment approach. ACPA titers may be useful to monitor the clinical activity of RA.

2. Innate immunity

A2.1 **ACTIVATED NEUTROPHILS ARE ABLE TO EFFICIENTLY PRODUCE INTERFERON-α AND RETAIN THIS CAPABILITY IN SYSTEMIC LUPUS ERYTHEMATOSUS AND RHEUMATOID ARTHRITIS PATIENTS**

[1]Dennis Lindau, [1]Armin Rabsteyn, [2]Julie Mussard, [2]Matthieu Ribon, [3]Ina Kötter, [4]Gosse Adema, [2]Marie-Christophe Boissier, [2,]*Patrice Decker. [1]*University of Tübingen, Institute for Cell Biology, Department of Immunology, Tübingen, Germany; [2]University of Paris 13, Sorbonne Paris Cité EA4222, Li2P, Bobigny, France; [3]University Hospital, Internal Medicine II, Tübingen, Germany; [4]Nijmegen Centre for Molecular Life Sciences, Department of Tumor Immunology, Nijmegen, The Netherlands*

*Corresponding and presenting author. E-mail: patrice.decker@univ-paris13.fr

Background and Objectives Neutrophils play a pivotal role in inflammation and contribute to systemic lupus erythematosus (SLE) and rheumatoid arthritis (RA) pathogenesis. Interferon (IFN)-α is involved in lupus development and might contribute locally to RA development. Activated plasmacytoid dendritic cells (pDC) are important producers of IFN-α but represent a minor cell population. On the other hand, neutrophils represent 50% of total blood leukocytes. Although neutrophils are not considered as IFN-α-producing cells, we have investigated whether they may produce this cytokine and the stimuli involved.

Materials and Methods PBMC and neutrophils were isolated from healthy individuals, SLE and RA patients. Mouse neutrophils were purified from the bone marrow. Cells were activated with different stimuli and IFN-α production/secretion was estimated by flow cytometry, ELISA and a bioassay. Neutrophil activation was verified by flow cytometry and ELISA. Gene expression was analysed by qRT-PCR. Neutrophil extracellular trap (NET) induction was estimated by confocal microscopy. Chromatin, a major autoantigen in SLE, was purified from calf thymus.

Results Isolated neutrophils produce IFN-α upon stimulation with Toll-like receptor (TLR) 9 and TLR7/8 agonists. IFN-α secretion by neutrophils was observed with neutrophils from both healthy donors and SLE and RA patients. Similar results were obtained with mouse neutrophils. IFN-α production by neutrophils was associated with IL-8, IL-6 and TNF-α secretion, CD66b up-regulation, ROS production and increased gene expression levels of IFN-α, IFN-β and IL-6. In low responders, PBMC sustain IFN-α secretion by neutrophils in co-cultures. Neutrophil priming is not required but GM-CSF acts synergistically with TLR9 agonists. Particularly, neutrophils respond to all types (A, B and C) of CpG-oligonucleotides. pDC are more efficient than neutrophils in producing IFN-α at the single cell level but this was largely compensated by the 200-fold excess of neutrophils in whole blood. Importantly, neutrophil-derived IFN-α was detected in response to free chromatin, a lupus autoantigen, and was associated with neutrophil extracellular trap (NET) formation (NETosis).

Conclusions Neutrophils represent an important source of IFN-α. IFN-α was detected at the mRNA and protein levels and in an active and secreted form. Both normal as well as SLE and RA neutrophils produce IFN-α in response to specific stimuli. Therefore, neutrophils represent also important targets for future therapies aiming at influencing IFN-α levels.

A2.2 **ACUTE SERUM AMYLOID A (A-SSA) INDUCES TIMP-1 IN DERMAL FIBROBLASTS THAT IS MEDIATED VIA A TLR-DEPENDENT AND FORMYL RECEPTOR-INDEPENDENT PATHWAY**

Steven O'Reilly, Rachel Cant, Marzena Ciechomska, Jacob M van Laar. *Musculoskeletal Research Group, Institute of Cellular Medicine, Faculty of Medicine, Newcastle University, UK*

Background and Objectives Systemic sclerosis (SSc) is an autoimmune connective tissue disorder characterised by inflammation and fibrosis. The fibrosis is due to an accumulation of extracellular matrix material due to enhanced deposition and/or reduced degradation. We and others have shown that abnormal production of TIMP-1 may contribute to fibrosis in SSc. Acute Serum Amyloid A (A-SAA) is an acute phase reactant and its levels correlate with ESR and CRP thus it is relevant in inflammatory conditions and has been demonstrated to be elevated in serum from SSc patients. The aim of this study was to determine the effects of A-SAA on dermal fibroblasts IL-6 and TIMP-1 secretion and its signalling mechanism downstream of ligation.

Materials and Methods Human healthy dermal fibroblasts from punch biopsies were cultured at low passage and then treated with recombinant A-SAA (10 ug/ml) for 24 hours. After the set time points the cells were harvested and the supernatant removed.