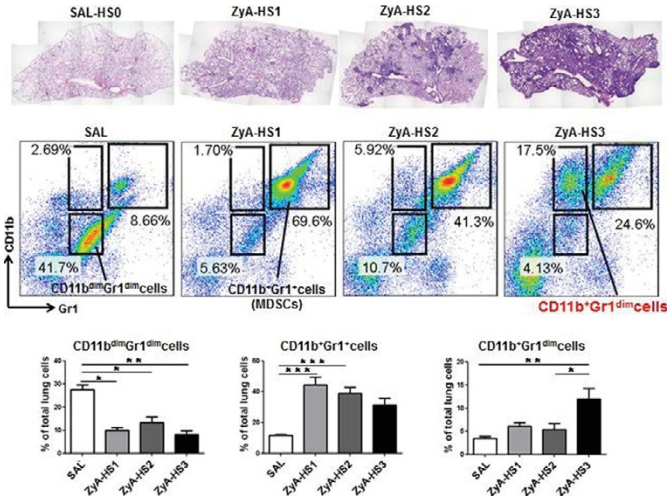


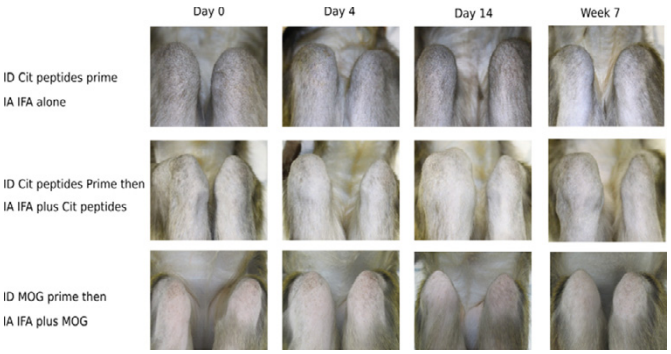
Figure 1

Disclosure of Interest: None declared
DOI: 10.1136/annrheumdis-2017-eular.1429

FRI0081 A MACAQUE MODEL OF RHEUMATOID ARTHRITIS BY IMMUNIZATION WITH CITRULLINATED PEPTIDES: LESSONS FOR THE HUMAN DISEASE

S. Bitoun[1], P. Roques[2], T. Larcher[3], G. Nocturne[1], C. Serguera[4], P. Chretien[5], G. Serre[6], R. Le Grand[7], X. Mariette[1]. *[1]Université Paris-Sud, Hôpitaux Universitaires Paris-Sud, INSERM U1184, le Kremlin Bicêtre; [2]Immunology of viral infections and autoimmune diseases, CEA, Fontenay aux Roses; [3]INRA UMR703 Veterinary School of Nantes, Nantes; [4]MIRCEN, CEA/INSERM, Fontenay aux Roses; [5]AP-HP, Hôpitaux Universitaires Paris-Sud, le Kremlin Bicêtre; [6]INSERM U1056 - Université de Toulouse Paul, Toulouse; [7]CEA - Université Paris Sud 11 - INSERM U1184, Fontenay aux Roses, France*

Background: Recent evolution in the understanding of rheumatoid arthritis (RA) mechanisms is the role of antibodies directed against citrullinated (cit) proteins (ACPAs). The shared epitope (SE) on the MHC class II is the main genetic risk factor of RA and favors presence of ACPAs. Mouse models dependent on cit peptides immunization require transgenic expression of the SE and are controversial. Non-human primates are ideal to study the interaction between ACPA and RA since 8% carry, similarly to humans, the SE called the H6 haplotype.
Objectives: The goal of this study was to develop a new animal model of RA based on immunization of genetically predisposed macaques against cit peptides to generate an ACPA-mediated model of arthritis.
Methods: Six macaques were intra dermally (ID) immunized with 4 peptides: vimentin (59–71) and (66–78), α fibrinogen (79–91) and aggrecan (89–103). H6 animals were immunized with either cit (n=2) or arginine (arg) (n=2) containing peptides. Two non H6 animals were immunized with cit peptides. These peptides are known to induce a T cell response in RA patients carrying the SE. T-cell response was assessed with Interferon γ ELISPOT and B-cell response by ELISA. An intra articular (IA) boost was done 30 weeks after initial immunization with either incomplete Freud's adjuvant (IFA) alone, IFA and cit peptides and IFA plus non relevant peptides.
Results: In the macaques, the T-cell response was specific to cit or arg peptides (depending on the peptides used for immunization). Surprisingly, the presence of the H6 epitope did not influence the response. Conversely the antibodies generated in response to the peptides were cross-reactive between the cit and arg peptides. Since no clinical response was observed, an IA boost was performed with the same 4 cit peptides and IFA adjuvant. This led to a prolonged neutrophil-rich mono-arthritis preferentially in H6 animals (Figure). Conversely, animals boosted with IFA alone only or with IFA plus myelin oligodendrocyte glycoprotein (MOG) peptides and previously immunized with MOG peptides presented with a transient mono-arthritis. Histological analysis revealed a local mononuclear infiltrate in one of the two animals that had prolonged knee monoarthritis. There was no clinical polyarthritis but 2 animals displayed synovial proliferation in 1 MCP and 1 MTP, respectively.
Conclusions: Immunization of macaques with cit peptides, then IA boost with the same cit peptides plus IFA, induced a prolonged monoarthritis. Shared epitope bearing did not restrict the T-cell response but seemed to favor the prolonged swelling after the IA boost. Neutrophil infiltration of the joint occurred similarly to what is seen in RA. Further use of neutrophil chemo-attractant might lead to a poly-articular disease. This macaque model of RA appears unique to study the events occurring during the pre-clinical phase of RA.



Disclosure of Interest: None declared
DOI: 10.1136/annrheumdis-2017-eular.2843

FRI0082 RAS SIGNALING INHIBITORS ATTENUATE ARTHRITIS IN ANIMAL MODELS OF RHEUMATOID ARTHRITIS BY DOWN MODULATING THE PATHOGENIC TH17 CELL RESPONSE

M. Zayoud[1,2], E. Vax[1], G. Elad Sfadia[3], V. Marcu-Malina[1], Y. Kloog[3], I. Goldstein[1,2,4]. *[1]Immunology Core Laboratory, Chaim Sheba Academic Medical Center, Ramat Gan; [2]Medicine, Sackler Faculty of Medicine, Tel Aviv University; [3]Faculty of Life Sciences, Tel Aviv University, Tel Aviv; [4]Rheumatology, Chaim Sheba Academic Medical Center, Ramat Gan, Israel*

Background: Ras-GTPases are vital for normal T cell activation, and downstream effectors of Ras include the MEK/ERK, PI3-kinase/AKT, mTOR/p70S6-kinase, and NF-kB pathways. Somatic mutations in *NRAS* cause an autoimmune lymphoproliferative disorder and T cells from Rheumatoid Arthritis (RA) patients exhibit perturbation of the Ras/MEK/ERK pathway. The small molecule Farnesylthiosalicylic acid (FTS) inhibits the interaction between Ras-GTPases and prenyl-binding chaperones vital for proper plasma membrane localization and downstream signaling [1]. Previous pre-clinical studies suggest that FTS has an immunomodulatory effect in various animal models of autoimmunity [2].
Objectives: To test in the Lewis rat adjuvant induced arthritis (AIA) and in the DBA/1 mouse collagen type-II induced arthritis (CIA) models the therapeutic immunomodulatory effect of FTS alone or combined with methotrexate (MTX).
Methods: Arthritis was induced in 8–12 week old male Lewis rats by complete Freund's adjuvant (CFA) injection and in male DBA/1 mice by collagen type-II (CII) immunization. Animals were treated prophylactically with once daily oral FTS (100 mg/kg); weekly i.p injection of MTX (0.5 mg/kg), oral FTS combined with MTX, or daily oral vehicle solution (0.5% carboxy methyl cellulose; CMC). Arthritis severity was scored daily from disease onset until study termination. In addition, we measured multiple disease- and drug-related immunological/molecular biomarkers.
Results: AIA severity was significantly reduced by FTS treatment compared to CMC controls (Figure 1A, $P<0.001$). Combining FTS and low dose MTX significantly increased its therapeutic efficacy compared to each drug alone (Figure 1A, $P<0.05$). FTS or FTS+MTX treatment also suppressed the upsurge in serum IL-17 and CRP compared to ailing controls. Global gene expression analysis of relevant splenic CD4+ T cells revealed that FTS is a potent inhibitor of pro-inflammatory and TH17 related gene networks. Next, our data from the mouse CIA model show that the therapeutic efficacy of FTS was non-inferior to MTX and it significantly reduced arthritis severity compared to controls (Figure 2, $P<0.001$). Importantly, FTS significantly inhibited the production of pathogenic anti-CII autoantibodies and upregulation of serum IL-6 and IL-17A compared to control arthritic mice. The in depth, multiplex, analysis of the effect of FTS on the T cell cytokine response to CII, revealed strong suppression of IL-22, IL-17, IL-9, GM-CSF and TNF production. Noteworthy, FTS therapy positively correlated with reduced Ras-GTP, p-ERK and p-AKT levels in splenic lymphocytes (drug related biomarkers).

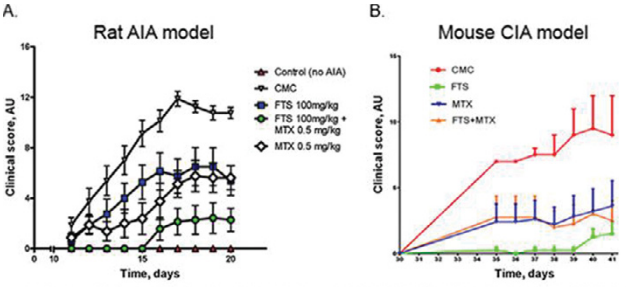


Figure 1. FTS attenuates arthritis severity in the (A) rat AIA and (B) mouse CIA models

Conclusions: FTS, a first-in-class oral selective Ras-GTPases inhibitor, exhibits a potent immunomodulatory effect in two classical murine model of arthritis, coupled with the inhibition of the TH17 response to relevant arthritogenic-antigens. Thus, Ras-signaling-blockade is a promising novel therapeutic approach for RA.

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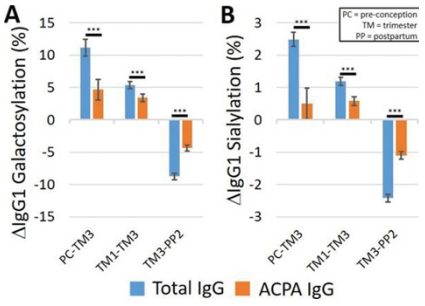
Disclosure of Interest: None declared
DOI: 10.1136/annrheumdis-2017-eular.1083

FRI0083 REDUCED INCREASE OF ACPA IGG-FC GALACTOSYLATION DURING PREGNANCY IN COMPARISON TO TOTAL IGG: AN EXPLANATION WHY AUTOANTIBODY POSITIVE RA-PATIENTS IMPROVE LESS DURING PREGNANCY?

A. Bondt[1,2,3], L. Hafkenscheid[3], D. Falck[2], T.M. Kuijper[1], Y. Rombouts[3], J.M. Hazes[1], M. Wuhrer[2], R.J. Dolhain[1]. *[1]Rheumatology, Erasmus University Medical Center, Rotterdam; [2]Center for Proteomics and Metabolomics; [3]Rheumatology, Leiden University Medical Center, Leiden, Netherlands*

Background: Rheumatoid arthritis (RA) disease activity (DAS28-CRP) improves less during pregnancy in autoantibody positive patients.[1] The most specific autoantibodies for RA are anti-citrullinated protein antibodies (ACPAs), which mainly occur as the immunoglobulin (Ig) G isotype. An association with DAS28-CRP and the pregnancy-associated improvement is well established for the Fc glycosylation of total IgG, in particular for galactosylation (Gal) and sialylation (SA).[2] The Fc glycosylation of ACPAs – mainly present as IgG – has been reported to be different from the total IgG Fc glycosylation.[3]
Objectives: We sought to determine whether the change in ACPA IgG glycosylation during pregnancy is different from that of total IgG, and whether this relates to the improvement of RA during pregnancy.
Methods: ACPA positive patient sera (*n*=152) were obtained within the framework of the PARA cohort, a prospective study designed to investigate pregnancy-associated improvement of RA. ACPA IgG was isolated using microscale affinity chromatography. Trypsin digested ACPA IgG was measured using nano-liquid chromatography mass spectrometry, and compared to total IgG.
Results: Pregnancy-associated changes in the levels of glycosylation were observed for all ACPA IgG subclasses. Pregnancy-associated glycosylation changes were less pronounced during pregnancy and after delivery in ACPA IgG (Gal +5%; SA +0.5%) compared to total IgG (Gal +11%; SA +2.5%; Figure 1), but – for total IgG – not different between ACPA+ and ACPA- patients. No association of the change in DAS28-CRP with the change in ACPA IgG or total IgG galactosylation was observed for ACPA+ patients, whereas a strong association of total IgG galactosylation was observed for ACPA- patients.



Conclusions: During pregnancy the increase in galactosylation of ACPA IgG was less pronounced than that of total IgG, whereas the increase in the galactosylation of total IgG was not different between ACPA+ and ACPA- patients. Since it is known that changes in IgG galactosylation are associated with improvement of RA during pregnancy and since ACPA is thought to be of pathogenic significance in RA, our data might provide an explanation why ACPA+ RA patients are less likely to improve during pregnancy.
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Acknowledgements: This project is funded by the Dutch Arthritis Foundation (NR 10–1-411) and by the European Union's Seventh Framework Program (FP7-Health-F5–2011) under grant agreement no°278535 (HighGlycan). We thank Dr Jan Wouter Drijfhout (LUMC, Leiden) for providing the CCP2 peptide.
Disclosure of Interest: None declared
DOI: 10.1136/annrheumdis-2017-eular.6229

FRI0084 THE TONSIL MICROBIOME IS INVOLVED IN RHEUMATOID ARTHRITIS

J. Li[1], J. He[1], K. Deng[2], J. Qin[2], Y. Jin[1], J. Chen[1], Z. Li[1]. *[1]Department of Rheumatology and Immunology, Peking University People's Hospital, Beijing; [2]Digital Microbiota Technology Co. Ltd., Shenzhen, China*

Background: Rheumatoid arthritis (RA) is a prevalent systemic autoimmune disease characterized by the production of autoantibodies[1]. The tonsil has been demonstrated to be a site of citrullination, and tonsillectomy has been reported to be a potential treatment of RA, suggesting the possibility that the tonsil could be a site of autoimmunity generation in RA[2,3]. The dysbiosis of gut microbiome and the associated host immune response has been implicated in the initiation and progression of RA[4–6]. However, there is no in-depth studies on the role of tonsil microbiota in RA. Thus, studies of the characteristics of tonsil microbiome in RA patients, the underlying mechanisms, as well as specific markers for the diagnosis and therapeutic evaluation for RA, are critical for the early diagnosis and prevention of RA.
Objectives: Therefore, we aimed to define the association of RA with tonsil microbiome as well as a microbial and metabolite profile that could predict disease status.
Methods: 16S rRNA gene sequencing was utilized on 220 tonsil swab samples (121 RA patients and 99 healthy controls) as well as 78 fecal samples (68 RA and 10 controls). Analysis of microbial taxa and metabolic pathway were performed to characterise and compare the tonsil microbiome of RA patients and healthy subjects
Results: Results showed that the tonsil harbored a unique microbiome relative to that present in the fecal samples. Patients with RA exhibited different tonsil microbiome from controls. A taxon-level analysis suggested that the relative abundance of 26 microbial clades were significantly altered, with 7 taxa increased and 19 taxa decreased in RA samples. Noticeably, we observed an expansion of rare microbial lineages as well as an alteration in microbial cladogenesis within RA patients. RA tonsil microbiota was associated with smoke, anti-peripheral factor, rheumatoid factors, disease duration and activity. Furthermore, we identified that 86 genes associated with bacterial metabolic pathway were enriched in RA tonsil microbiome.
Conclusions: Our results demonstrated that the RA tonsil microbiome differs from that of healthy controls, which correlates with systemic autoimmune changes and may potentially drives initiation of RA.
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Disclosure of Interest: None declared
DOI: 10.1136/annrheumdis-2017-eular.4707

FRIDAY, 16 JUNE 2017

Rheumatoid arthritis - prognosis, predictors and outcome

FRI0085 NUMBER OF PEPTIDE-SPECIFIC ANTI-CITRULLINATED PEPTIDE ANTIBODIES IN SYNOVIAL FLUID AND IN SYNOVIAL FLUID IMMUNE COMPLEXES ASSOCIATE WITH DEGREE OF RADIOLOGICAL DESTRUCTION AND RESPONSE TO TRIAMCINOLONE HEXACETONIDE FOR KNEE SYNOVITIS IN RHEUMATOID ARTHRITIS

A. Sohrabian[1], L. Mathsson Alm[1,2], M. Hansson[3], J. Lysholm[4], M. Cornillet[5], A. Knight[6], K. Skriner[7], G. Serre[5], A. Larsson[8], T. Weitoft[9], J. Rönnelid[1]. *[1]Department of Immunology, Genetics and Pathology, Uppsala university; [2]ImmunoDiagnostic Division, Thermo Fisher Scientific, Uppsala; [3]Rheumatology Unit, Department of Medicine, Karolinska Institutet, Stockholm; [4]Clinic of Rheumatology, Falun Hospital, Falun, Sweden; [5]Epithelial Differentiation and Rheumatoid Autoimmunity, U 1056 Inserm Toulouse University, Toulouse, France; [6]Rheumatology, Department of Medical Sciences, Uppsala university, Uppsala, Sweden; [7]Department of Medicine, Charité University Hospital, Berlin, Germany; [8]Section of Clinical Chemistry, Department of Medical Sciences, Uppsala university, Uppsala; [9]Section of Rheumatology, Center for Research and Development, Uppsala university, Gävle, Sweden*

Background: We have described a planar microarray for the determination