**Results:** The agglutination mediator that was generated is based on a single-chain antibody fragment that binds to glycophorin A,[3] one of the major surface proteins of erythrocytes. It is conjugated to a citrullinated peptide that is efficiently recognized by ACPA. In the presence of erythrocytes and ACPA the mediator induces agglutination of the erythrocytes, which can be detected by the naked eye. The addition of the mediator resulted in detectable agglutination in 64% percent of the RA patient samples. Agglutination correlated well with the results obtained with a commercial anti-CCP2 ELISA for ACPA detection. Efficient agglutination was observed with only 7% of the PsA samples. No correlation with rheumatoid factor levels was observed.
**Conclusion:** An ACPA-dependent hemagglutination mediator was generated. This agglutination mediator allows the rapid and efficient detection of ACPA by hemagglutination in human blood samples.
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## AB0050 EVALUATION OF THE ASSOCIATION BETWEEN CIRCULATING NON-CODING RNA CIRC_0005567 AND DISEASE ACTIVITY IN PATIENTS WITH RHEUMATOID ARTHRITIS AND ITS BIOLOGICAL FUNCTION IN THE CELL LINE MODEL

**Keywords:** Genetics/Epigenetics, Biomarkers, Rheumatoid arthritis

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**Background:** Rheumatoid arthritis (RA) is a chronic, autoimmune disease that leads to chronic inflammation of synovial tissue, ultimately causing joint damage, disability and premature mortality. Disease affects about 1% of the global population. Circular RNAs (circRNAs) are non-coding molecules and are generated through back splicing, during which the 5 ‘ and 3′ ends are covalently joined. Consequently, the lack of free ends makes them stable and resistant to exonucleases, and they become more suitable biomarkers compared with linear RNAs such as microRNAs (miRs) or long non-coding RNAs. CircRNAs are still poorly understood molecules in relation to RA pathogenesis.
**Objectives:** The aim of the study was to find an association between circ_0005567 plasma levels and disease activity in RA patients and to evaluate its molecular function.
**Methods:** A total of 66 individuals, 39 RA patients, and 27 healthy controls. RA patients were selected based on disease activity and RA patients with high disease activity (DAS28 >5,1) and remission (≤2,6) were included. RNA was extracted from plasma and quantitative real-time PCR was used to analyze concentration of circ_0005567. The SW982 cell line was cultured in Dulbecco’s Modified Eagle’s Medium and supplemented with 10% fetal bovine serum. The cell line was grown at 37 °C, in a humidified atmosphere, with 5% CO2. The circ_0005567 silencing was performed by designed small interfered RNAs (siRNAs). Bioinformatic analysis was applied to select the miRs and mRNAs that may interact with circ_0005567. The expression levels of microRNAs were evaluated, subsequently the expression levels of selected mRNAs were performed. The expression levels of miRs and mRNA were evaluated at three time points: after 24h, 48h, and 72h.
**Results:** The expression level for circ_0005567 in RA patients was elevated compared to the control group (mean±SD; 173.6±123.8 vs 108.9±81.03, p=0.017). The molecule also showed significant differences in disease activity. Patients with high disease activity had about 70% higher concentration of circ_0005567 than the control group (186.99 vs 108.9, p=0.015). In the cell line model we found an association between silencing circ_0005567 and elevated miR-194 concentration and elevated concentration of three mRNAs: *KPNA1, HBEGF, TLN2.*
**Conclusion:** The expression of circ_0005567 was related to genes that are significant for the pathogenesis of RA. For example, *HBEGF* gene expression in fibroblasts plays a role in the remission of rheumatoid arthritis, and *KPNA1* was reported as seed gene related to RA. Due to the fact that circRNAs are stable and resistant to degradation, plasma concentration of circRNAs may be a new potential epigenetic markers of RA and disease activity.
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## AB0051 DESCRIPTION OF PULMONARY INVOLVEMENT IN THE RAT ADJUVANT-INDUCED ARTHRITIS MODEL

**Keywords:** Rheumatoid arthritis, Animal models, Lungs

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**Background:** Interstitial lung disease (ILD) is detected until 60% of patients with rheumatoid arthritis (RA), but is clinically significant in around 10% of cases. It is one of the leading causes of disease and death in RA patients. High levels of anti-cyclic citrullinated peptide (CCP) autoantibodies are associated with co-occurrence of pulmonary disease and RA [1]. Moreover, the pulmonary fibrosis marker mucine (MUC-1) was described to be a predictive indicator for ILD [2]. The adjuvant-induced arthritis (AIA) model is commonly used to investigate arthritic diseases due to some pathophysiological similarities to human arthritis [3].
**Objectives:** The first objective of this study was to investigate the pulmonary involvement occurring in the rat AIA model. The second objective was to investigate the presence of potential biomarkers such as CCP and MUC-1.
**Methods:** This study was performed from lungs from 64 AIA rats and 6 control rats without arthritis. Lungs were collected to perform histological and immunological analyses. Pulmonary disease was measured with a score based on the percentage of lung damage and the thickness of the alveolar walls (0: absent, 1: mild, 2: moderate, or 3: severe). CCP and MUC-1 immunostaining was also performed.
**Results:** A nonspecific interstitial lung disease was detected in the lungs of 80% of AIA rats, whereas only half control rats developed it. A heterogeneous percentage of lung damage was observed, which correlated with the thickness of the alveolar walls but not with arthritis severity. Granulomas were observed in the most affected lung tissues, with a slight fibrosis at their periphery. The number of CCP markings correlated positively with the percentage of lung damage in AIA rats. MUC-1 was found homogeneously in the lung tissue of control and AIA rats, but more prominently in the periphery of granulomas in AIA rats.
**Conclusion:** We have described the pulmonary involvement in the AIA rat model, which developed the extra-articular complication similar to RA-ILD. We identified CCP and MUC-1 as potential biomarkers of this pulmonary involvement. Thus, the lung may be a site of initiation of CCP immunity. The AIA model will then allow a better understanding of RA-ILD, which may lead to earlier diagnosis and the potential development of new targeted therapies.
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