mRNA showed no significant difference in fold change between SpA patients and healthy controls.

**Table 1. Results of DNA promoter methylation and previously performed gene expression analysis in juvenile spondyloarthritis patients.**

<table>
<thead>
<tr>
<th>GENES</th>
<th>Fold Enrichment of Immunoprecipitated DNA</th>
<th>Fold Change</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patients</td>
<td>Controls (n=19)</td>
<td>p</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TLR4</td>
<td>0.5759</td>
<td>-3.91</td>
</tr>
<tr>
<td>NLRR3</td>
<td>0.0220</td>
<td>-5.00</td>
</tr>
<tr>
<td>CXCRR4</td>
<td>0.2658</td>
<td>-1.87</td>
</tr>
<tr>
<td>PTPRH2</td>
<td>0.3768</td>
<td>-2.63</td>
</tr>
</tbody>
</table>

**Conclusions:** Our study indicated epigenetic modifications are probably responsible for some of the expression alterations in SpA patients in the initial phase of the disease. Since NLRR3 has a crucial role in inﬂammatory assembly and inﬂammosomes have been shown to shape microbiota, it is reasonable to assume dysbiosis in SpA patients can at least partially be explained by reduced NLRR3 expression due to hypermethylation, stressing for the first time the epigenetic contribution to SpA pathophysiology. While this is not clear if these epigenetic alterations are caused by genetic mutations in epigenetic factors or exposure to certain environmental factors that modulate the occurrence of aberrant epigenetic signatures, the discovery of DNA methylation-based signature of the NLRR3 gene could have important implications in addressing intrinsic and extrinsic contribution to SpA pathophysiology, whereas the possibility of reverting epigenetic modifications opens new prospects for treating other components of this complex disease.

**REFERENCE:**


**FR0004**

**THE RELATIONSHIP BETWEEN JUVENILE SYSTEMIC LUPUS ERYTHEMATOSUS AND THE TRANSCRIPTION FACTORS NF-KAPPAB AND PPAR-GAMMA**

S. Sahin1, S. Durmus2, A. Ardovci, K. Barut1, R. Gelisgen2, H. Uzun2, O. Kasapcoglu1, 1Pediatric Rheumatology, 2Department of Biochemistry, Istanbul University, Cerrahpasa Medical School, Istanbul, Turkey

**Background:** Systemic lupus erythematosus (SLE) is an autoimmune disease characterised by high-levels of autoantibodies mainly targeting nuclear antigens and loss of self-tolerance. Peroxisome-proliferator activated receptor gamma (PPARγ) and nuclear factor-kappa beta (NF-κB) are transcription factors, which, within normal levels, have shown to be crucial in immunomodulation namely, activation and development of normal lymphocytes, negative and positive selection of T and B cells. High-levels of NF-κB has inflammatory properties such as release of autoreactive T cells. On the contrary, PPARγ has anti-inflammatory effects, which has been demonstrated to be effective when used early in prevention of disease in murine models of systemic lupus erythematosus.

**Objectives:** Herein, we investigated whether NF-κB and PPARγ could exert opposite effects in the immune response and the possible implications in immunomodulation of juvenile systemic lupus erythematosus.

**Methods:** Serum NF-κB and PPARγ levels were measured in 42 juvenile systemic lupus erythematosus. In addition, 19 juvenile systemic sclerosis and 25 age-matched healthy children were selected for patient control and healthy control, respectively. We have also assessed the relation of these transcription factors with disease activity and anti-ds DNA.

**Results:** The control group did not differ from the juvenile SLE and juvenile systemic sclerosis patients for age (p>0.05). According to our study, serum NF-κB levels of juvenile SLE and juvenile systemic sclerosis patients were significantly higher (1.87±1.0 and 2.17±1.0 versus 1.25±0.7), while serum PPARγ levels were significantly lower than that of healthy controls (1.52±0.5 versus 2.03±0.9). The difference was not significant between juvenile systemic lupus erythematosus and juvenile systemic sclerosis. In patients with juvenile systemic sclerosis serum NF-κB levels negatively correlated with serum PPARγ levels (R= −0.49; p=0.032); however, this relationship was not observed in juvenile SLE patients and healthy controls.

**Conclusions:** Increased serum NF-κB levels represent upregulated signalling cascades, so it is associated with increased levels of pro-inflammatory cytokines. Since juvenile systemic sclerosis and juvenile systemic lupus erythematosus are autoimmune diseases, patients had high levels of NF-κB and low levels of PPAR than controls, as expected. Previous studies revealed that PPARγ activation inhibits NF-κB transcriptional activity. Correlation results in juvenile systemic sclerosis cohort are compatible with this finding, however not in juvenile systemic lupus erythematosus patients. This could be due to the limited number of patients. Further studies with large number of patients are needed to better elucidate the implication of these transcription factors in therapeutic pathways.

**REFERENCES:**


**FR0005**

**GRANULOCYTE MACROPHAGE COLONY STIMULATING FACTOR IS SECRETED AT HIGHER LEVELS FROM STIMULATED MONOCYTE-DERIVED MACROPHAGES FROM PATIENTS WITH ENTHESITIS RELATED ARTHRITIS AND IS SIGNIFICANTLY ENHANCED BY THE UNFOLDED PROTEIN RESPONSE**

C. Fischer1,2, D. Elefteriou1, D. Seri1,2, Y. Ioannou1. 1Arthritis Research UK Centre for Adolescent Rheumatology, University College London; 2National Institute for Health Research University College London Hospitals Biomedical Research Centre, University College London and University College London Hospital, London, UK

**Background:** Enthesitis related arthritis (ERA) is a subtype of juvenile idiopathic arthritis exhibiting many similarities to the adult spondyloarthopathies (SpA). The innate immune system and intracellular stress responses, including the unfolded protein response (UPR), have been implicated in the pathogenesis of SpA, Granulocyte macrophage colony stimulating factor (GMCSF), as well as being a haematopoietic growth factor, plays a central role in regulating innate immunity and has been shown to be crucial in immunomodulation namely, activation and development of normal lymphocytes, negative and positive selection of T and B cells. High-levels of NF-κB has inflammatory properties such as release of autoreactive T cells. On the contrary, PPARγ has anti-inflammatory effects, which has been demonstrated to be effective when used early in prevention of disease in murine models of systemic lupus erythematosus.

**Objectives:** To compare levels of GMCSF produced by monocyte-derived macrophages (MDMs) from patients with ERA and healthy controls and to observe the effect of inducing the UPR on those levels.

**Methods:** Peripheral blood monocytes were isolated from 39 patients with ERA and healthy controls and differentiated in vitro with macrophage-colony-stimulating factor. Cells were treated with interferon gamma for 24 hours to upregulate HLA B, washed and then stimulated with lipopolysaccharide (LPS) alone (50 ng/mL) or LPS and thapsigargin (TM) (5 μM), an inducer of the unfolded protein response. GMCSF was measured from the cell culture supernatants after 24 hours culture by luminescent assay.

**Results:** Levels of GMCSF at baseline were similar in patients and healthy controls [median 121.3 pg/mL (IQR 96.6–194.0 pg/mL) vs 157.1 pg/mL (124.2–203.3 pg/mL), p=0.1]. However, with LPS stimulation, MDMs from patients secreted significantly higher levels of GMCSF [median 1853 pg/mL (IQR 1206–3061 pg/mL) vs 1342 pg/mL (IQR 713.3–1797 pg/mL), p=0.0057]. On stimulation with TM in addition to LPS, GMCSF production was further enhanced in both patients and healthy controls [median 9027 pg/mL (IQR 4746–13961 pg/mL) vs 3834 pg/mL (IQR 1603–9158 pg/mL) and remained significantly higher in patients (p=0.0096). To investigate the effect of the UPR, fold change in GMCSF was calculated for each sample between MDMs stimulated with LPS alone and MDMs stimulated with both LPS and TM. Median fold change in patients was 3.95 (IQR 1.54–5.47) and 2.36 (IQR 0.49–4.69) in healthy controls. Interestingly, MDMs from patients who were HLA B27 positive exhibited significantly higher median fold change in GMCSF with UPR induction compared to HLA B27 negative patients [4.14 (IQR 2.22–8.60) vs 1.33 (IQR 0.36–3.91), p=0.0098]. No associations were seen with different treatment regimes in the patient group.

**Conclusions:** MDMs from patients with ERA produce significantly higher levels of GMCSF after stimulation compared to healthy controls and this is further enhanced by the UPR, especially in HLA B27 positive patients. These results potentially implicate GMCSF in the pathogenesis of ERA and further support the concept of GMCSF as a novel target for treatment in certain subgroups of patients.

**REFERENCE:**