Pathogenic insights transforming the treatment of Sjögren’s and SLE 2020 and beyond

**OP0042**

THE TRANSCRIPTOME OF PAIRED MINOR AND MAJOR SALIVARY GLAND TISSUE IN PATIENTS WITH PRIMARY SJÖGREN’S SYNDROME: TWO OF A KIND?

G. M. Verstappen1, L. Gao2, S. A. Pringle1, S. C. Liefers2, B. Van der Vegt2, V. Patel1, S. Hu2, S. Mukherjee2, A. Vissink3, H. Bootsma1, F. G. M. Kroese1,1 University of Groningen, University Medical Center Groningen, Rheumatology and Clinical Immunology, Groningen, Netherlands; 2Bristol-Myers Squibb, Princeton, United States of America; 3University of Groningen, University Medical Center Groningen, Oral and Maxillofacial Surgery, Groningen, Netherlands; 4University of Groningen, University Medical Center Groningen, Pathology and Medical Biology, Groningen, Netherlands

**Background:** In patients with primary Sjögren’s syndrome (pSS), both minor and major salivary gland tissue is targets of the disease. Infiltration of salivary gland by immune cells is characteristic for pSS. However, significant inter- and intra-individual variation exists in the size and composition of the infiltrates. Potential differences between minor and major salivary glands in immune cell presence and inflammatory pathway activation are unclear. This knowledge is essential for clinical trial design and precision therapy.

**Objectives:** To compare the transcriptome of paired labial and parotid salivary gland tissue from patients with pSS and non-SS sicca controls.

**Methods:** Thirty-nine pSS patients and 20 age- and sex-matched non-SS sicca controls, who participated in a prospective diagnostic cohort[1], were included. All pSS patients fulfilled 2016 ACR-EULAR criteria. RNA was isolated from formalin-fixed, paraffin-embedded labial and parotid gland tissue sections from the same individuals. Complementary DNA libraries were prepared and sequenced. Biopsies with evidence of sclerosing chronic sialoadenitis or mucosa-associated lymphoid tissue lymphoma were excluded in the current study. The transcriptome of labial and parotid gland tissue from pSS patients based on gene expression. When comparing the labial and parotid gland transcriptome, resp. 798 and 1461 DEGs (FDR-adjusted p-value<0.05, log2 fold change >1) were identified between groups I and IV. The top differentially regulated genes were mostly related to T and B cells. C2CL13, CCR6, MS4A1 (CD20), FCRL4 and DAZL were among the genes with the highest positive fold change in both glands of biopsy-positive pSS patients. Overall, there was a moderate to strong correlation between fold changes in labial and parotid glands (R²=0.58, p-value<0.0001). Between biopsy-negative and biopsy-positive pSS patients, overall, there was a moderate to strong correlation between fold changes in labial and parotid glands (R²=0.58, p-value<0.0001).

**Conclusion:** The transcriptome of labial and parotid gland tissue from pSS patients with a positive biopsy is overall comparable, while salivary gland tissue from biopsy-negative pSS patients shows a comparable gene expression profile to non-SS sicca controls. These results indicate that different treatment strategies may be necessary for biopsy-negative and biopsy-positive pSS patients.

**References:**

**Disclosure of Interests:** Gwenwy M. Verstappen: None declared, Lu Gao: Employee of: BMS, Sumanta Mukherjee: None declared, Sivas C. Liefers: None declared, Bert van der Vegt Consultant of: Advisory board member for Philips and Visiopharm., Vishal Patel Employee of: BMS, Sarah Hu Shareholder of: Bristol-Myers Squibb, Employee of: Bristol-Myers Squibb, Sumanta Mukherjee Employee of: BMS, Arjan Vissink: None declared, Hendrika Bootsma Grant/ research support from: Unrestricted grants from Bristol-Myers Squibb and Roche, Consultant of: Consultant for Bristol-Myers Squibb, Roche, Novartis, Medimmune, Union Chimique Belge, Speakers bureau: Speaker for Bristol-Myers Squibb and Novartis., Frans G.M. Kroese Grant/research support from: Unrestricted grant from Bristol-Myers Squibb, Consultant of: Consultant for Bristol-Myers Squibb, Speakers bureau: Speaker for Bristol-Myers Squibb, Roche and Janssen-Cilag

Background: There is an ongoing effort to elucidate the molecular pathways that are key to kidney injury in lupus nephritis (LN). One approach is to study the transcriptome utilising kidney tissue obtained during diagnostic renal biopsy [1]. In clinical practice the most common tissue that is surplus to diagnostic requirements is formalin-fixed paraffin-embedded (FFPE) tissue. However, due to RNA degradation, transcriptomic analysis has been sub-optimal and challenging using standard procedures. The NanoString technology platform has the advantage that reliable detection of transcripts can be achieved even with degraded RNA. In this study we explored the utility of NanoString technology in identifying transcripts in RNA isolated from archival FFPE kidney biopsy sections in a cohort of patients with LN.

Methods: We utilised well defined Class III (n=11); Class IV (n=22) and Class V (n=24) LN FFPE kidney biopsy specimens from female patients attending the Imperial College Healthcare NHS Trust. We excluded biopsies with mixed lesions or chronic kidney disease (n=14) disease were used as controls. Six 10 micron thick sections were obtained from each biopsy and RNA isolated using the Qiagen RNeasy FFPE Kit. 100 micrograms of RNA was used for the detection of transcripts in formalin-fixed paraffin-embedded Lupus Nephritis kidney biopsy tissue.

Objectives: To explore the utility of the NanoString platform in elucidating a renal transcriptomic signature in formalin-fixed paraffin-embedded Lupus Nephritis kidney biopsy tissue.

Results: Transcriptomic data passing NanoString nSolver quality control metrics was obtained from all sections. Notably sections included biopsies up to 16 years old. We detected 800 transcripts, including 40 reference genes. Transcript analysis was performed according to manufacturer’s instructions using the NanoString nSolver software. When analysing differential gene expression (DGE) we used Benjamini-Hochberg adjustment to account for multiple testing. The threshold for statistical significance was an adjusted P value of 0.05 (5% false discovery rate).

DGE analysis was performed on all sections with positive control for each class (Class III: baseline (week 0), wk 12, and wk 24) from SLE pts (n=270) and 50 sex- and age-matched controls. Samples were analyzed for: IL-2, IL-3, IL-5, IL-6, IL-10, IL-17A, IL-21, IL-12/23p40, IL-12p70, GM-CSF, IFN-α, IFN-γ, IL-12, and IL-23.

The objectives of the current study were: 1) to examine baseline serum cytokines in the JAHH phase 2 clinical trial for correlations with clinical or immunologic assessments; 2) to determine if changes in serum cytokine levels were associated with bari treatment.

Methods: Pts enrolled in the JAHH phase 2 trial received daily treatment with PBO, baricitinib 2 mg, or baricitinib 4 mg through week 24. Serum samples were collected at baseline (week wk 0), wk 12, and wk 24 from SLE pts (n=270) and 50 sex- and age-matched controls. Samples were analyzed for: IL-2, IL-3, IL-5, IL-6, IL-10, IL-17A, IL-21, IL-12/23p40, IL-12p70, GM-CSF, IFN-α, IFN-γ using ultrasensitive quantitative assays. IFN gene signature, autoantibodies, C3 and C4 were measured as previously described [1].

Conclusions: We had successfully identified transcriptomic signatures in RNA samples derived from a relatively large cohort of FFPE LN samples. Consistent with published reports we could detect a type I IFN signature in the LN kidney tissue [1]. Consistent with a recent study [1], we detected increased expression of OPN (osteopontin) and FN1 (fibronectin-1) in proliferative (Class III and IV) but not Class V LN. We are now performing clinical correlations to determine if the differentially expressed transcripts are clinically informative.

References:

**Disclosure of Interests:** Alyssa C. Gilmore: None declared, Hannah Wilson: None declared, Tom Cairns: None declared, Mariona Botto: None declared, Liz Lightstone Grant/research support from: Roche - ended 2018, Consultant of: GSK, Aurinia, Pfizer, Achillion, Speakers bureau: Alexion, Ian N. Bruce Grant/research support from: Genzyme Sanofi, GSK, and UCB, Consultant of: Eli Lilly, Azab-Zenecha, UCB, Itto, and Merck Serono, Speakers bureau: UCB, Terry Cook Grant/research support from: Achillion funding for natural history study on C3 glomerulopathy, Consultant of: Scientific consultant to Apellis, Alexion, GSK, Speakers bureau: Alexeon, David Pickering Grant/research support from: Funding for investigation of therapeutic compounds in pre-clinical models of complement-mediated kidney disease; Achillion funding for natural history study on C3 glomerulopathy, Consultant of: Scientific Advisor for Alexion, Achillion, Apellis

DOI: 10.1136/annrheumdis-2020-eular.1169

**Disclosure of Interests:** Alyssa C. Gilmore: None declared, Hannah Wilson: None declared, Tom Cairns: None declared, Mariona Botto: None declared, Liz Lightstone Grant/research support from: Roche - ended 2018, Consultant of: GSK, Aurinia, Pfizer, Achillion, Speakers bureau: Alexion, Ian N. Bruce Grant/research support from: Genzyme Sanofi, GSK, and UCB, Consultant of: Eli Lilly, Azab-Zenecha, UCB, Itto, and Merck Serono, Speakers bureau: UCB, Terry Cook Grant/research support from: Achillion funding for natural history study on C3 glomerulopathy, Consultant of: Scientific consultant to Apellis, Alexion, GSK, Speakers bureau: Alexeon, David Pickering Grant/research support from: Funding for investigation of therapeutic compounds in pre-clinical models of complement-mediated kidney disease; Achillion funding for natural history study on C3 glomerulopathy, Consultant of: Scientific Advisor for Alexion, Achillion, Apellis

DOI: 10.1136/annrheumdis-2020-eular.2394