RNA Sequencing Detection of Gene Dysregulation in B Cells Sorted from Salivary Gland Tissue and from Peripheral Blood Reveals New Pathways Involved in Primary Sjögren's Syndrome Pathophysiology

Abstract # 7

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Background/Purpose:
Primary Sjögren's syndrome (pSS) is a chronic auto-immune disorder characterized by lymphocytic infiltrates and destruction of the salivary glands. Chronic B cell activation, the secretion of autoantibodies and the critical role of BAFF have been demonstrated. However, mechanisms leading to B cells dysregulation remain partially understood.

The objective of this study was to establish transcriptomic maps of the B cells sorted from the salivary glands and from blood in pSS patients and controls.

Methods:
Patients had pSS according to 2016 EULAR/ACR criteria and controls had sicca symptoms without any antibodies and with normal salivary gland biopsy. B cells were sorted from salivary gland biopsies and from blood. Total RNASeq profiling was performed using MiSeq (Illumina). Statistical analysis (DESeq2) identified differentially expressed genes between pSS and controls in B cells sorted from salivary glands (9 pSS patients and 4 controls), from blood (16 pSS patients and 7 controls); and between B cells sorted from salivary glands and blood in the same patients (4 pSS patients). Functional enrichment analysis was performed using Ingenuity Pathway Analysis software.

Results:
The pSS vs controls comparison in B cells sorted from salivary glands identified up-regulated genes involved in activation of B cells including CD48, CD22 and CD40. TLR10, which is involved in innate immunity was also up-regulated in pSS. The analysis of the non-coding expressed RNAs showed an up-regulation of Mir155 which is essential for B cell differentiation and antibody production (**Table 1A**).

In blood B cells, TLR7 and the downstream signaling molecule IRF7 were up-regulated in pSS. Additionally, IL-6 which is involved in B cells growth was up-regulated (**Table 1B**). Enrichment analysis highlighted EIF2 signaling pathway, interferon (IFN) signaling pathway and role of JAK in IFN signaling (**Table 2**).

The paired comparison between B cells from salivary glands and from blood identified up-regulated genes including CD138, a plasma cell marker, IL-6, TLR5 and IFN induced genes (**Table 1C**). As non-coding RNA, Mir155 was also up-regulated.

The confirmation by qPCR method of these results is ongoing.

Tables available online

Conclusion:
This study allowed to explore the mechanisms that support B cell activation in pSS focusing on tissue resident and circulating cells. Our data confirmed the B cell activation and differentiation through several markers including CD40, CD22, CD48, CD138 and highlighted the role of innate immunity with the TLRs and key pathways including IFN and JAK signaling. Precise understanding of these dysregulation should offer development of new targeted therapeutic perspectives for patients.