Flow Cytometric Immunophenotyping of Salivary Glands in Primary Sjögren's Syndrome

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Background

Primary Sjögren's Syndrome (PSS) is a common autoimmune disease of unknown aetiology. It is characterised by inflammatory infiltration of exocrine glands, development of a sicca syndrome and a 20-fold increase in the risk of developing lymphoma. Standard pathological evaluation is based on a lymphocyte 'focus score' but little is known about the composition of the lymphoid infiltrate or its relationship to disease markers such as autoantibodies and the risk of lymphoma. The aim of the study was to use flow cytometry to characterise the lymphoid infiltrate in more detail.

Methods

Salivary glands were collected from 103 subjects attended the Newcastle Sjogren's clinic who had undergone minor salivary gland biopsy as part of the diagnostic investigations which also include testing for anti-SSA/SSB antibodies, Schirmer's tests and unstimulated oral salivary flow. 70 with confirmed PSS, 15 with potential or early-stage PSS and 18 with non-SS. Salivary glands were digested in collagenase for 3 hours and sort-analysed using a BD Biosciences FACSFusion flow cytometer. Sorted cells from 6 patients were giemsa stained to observe cell morphology. All subjects have given their written informed consent according to the principles of Helsinki and the project has received local REC approval.

Results & Discussion

Our data show that it is relatively easy to identify multiple lymphocyte populations from salivary glands by flow cytometry. Salivary glands contain CD19+ B cells, CD19+CD38+ plasmablasts, CD19-CD38+ plasma cells, and predominantly central memory CD4+ and CD8+ T cells. In PSS salivary glands, the total number of lymphocytes is increased by up to 10-fold with a skewing towards higher numbers of B lineage cells. In particular there was a striking increase in the number of CD19-CD38+ cells with marked kappa light chain restriction in the most advanced cases. By morphology these cells had the appearance of plasma cells and included binucleate and vacuolated forms traditionally only associated with plasma cell dyscrasias.

Conclusion

Flow cytometry reveals clear correlations with focus score in PSS patients. The dominant population in PSS salivary glands is a previously uncharacterised tissue plasma cell with light chain restriction consistent with that observed in patient plasma. Further investigations need to be performed to define potentially pathogenic populations.