

## The active immunological profile in patients with primary Sjögren's syndrome is restricted to typically encountered autoantibodies

Sirs,

Primary Sjögren's syndrome (pSS) patients can be distinguished by combination of sicca symptoms such as xerostomia, xerophthalmia, salivary gland (SG) biopsy showing focal infiltration by lymphocytes and the detection of autoantibodies (Ab) directed against sicca syndrome autoantigens (SSA)/Ro and SSB/La (1). However, pSS patients are also characterised by a broad spectrum of systemic manifestations and multiple Ab, of which anti-nuclear Ab (ANA  $\geq 1:320$ ) and rheumatoid factors (RF) are the most frequently detected (2, 3).

Considering that a serum-based approach combining different Ab would be meaningful for characterisation of pSS patient with an active immunological profile, we first decided to conduct an analysis to evaluate the performance of conventional Ab including ANA on HEp-2 cells, anti-SSA/Ro Ab (native 52+60kDa), anti-SSB/La Ab, and IgM RF in a well-characterised population of pSS patients ( $n=113$ ; 92.9% were female) who had benefited from a comprehensive clinical, histological and radiological assessment (4).

The analysis for typical Ab showed ANA (71.7% at 1:320), anti-SSA/Ro Ab (61.1%), anti-SSB/La Ab (31.0%) and IgM RF (44.2%). In addition, and since we observed interrelationships between Ab that are suspected to have prognostic value in pSS (5-8), we further distinguished two groups of patients based on the triple association ANA+SSA+RF to characterise patients with an active immunological profile (Fig. 1A). Patients with the triple Ab association were referred to as group I ( $n=46$ ) in contrast to group II patients ( $n=67$ ) that possessed up to 2 Abs. Group I patients were characterised by a younger age at diagnosis ( $46.7 \pm 14.8$  years in group I vs.  $52.7 \pm 12.4$  years in group II,  $p=0.02$  data not shown). Moreover, and as presented in Figure 1B, SG involvement characterised patients from group I with regards to abnormal SG enlargement (odds ratio (OR)=2.6 CI95% 1.1-6.3,  $p=0.04$ ) and abnormal major SG ultrasonography (OR=22.8 CI95% 4.9-106,  $p<10^{-4}$ ) as recently established (9, 10). Those patients from group I more often had dense SG lymphocyte infiltration using grade 3 Chisholm-Mason scale (OR=3.5 CI95% 1.1-11.1;  $p=0.04$ ), hypergammaglobulinaemia over 20g/L (OR=6.9 CI95% 1.7-27.7,  $p=0.007$ ), and a higher erythrocyte sedimentation rate ( $29.2 \pm 23.6$  in group I vs.  $18.2 \pm 20.0$ ;  $p=0.01$ ). In contrast, no differences were observed with regards to clinical manifestations with the exception of interstitial pulmonary involvement (OR=3.6 CI95% 1.4-9.4,  $p=0.008$ ).

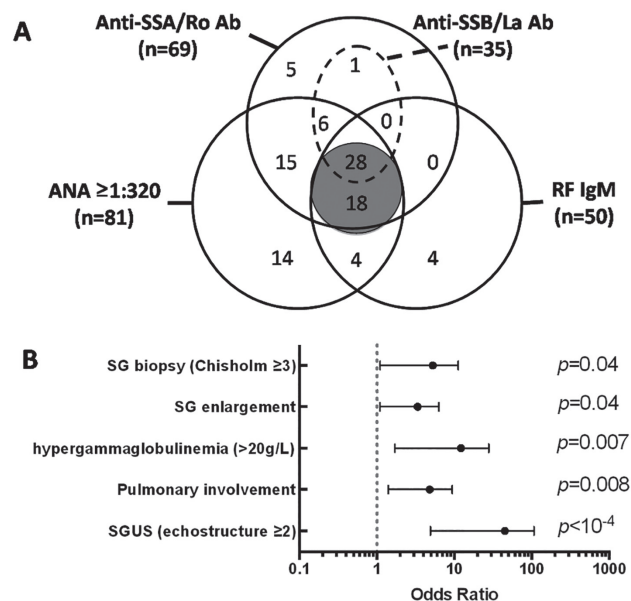
Then, later, the presence of atypical Ab was further explored in 44 pSS patients from the cohort using a new automated chemilumi-

Fig. 1. Interrelations between anti-sicca syndrome (SSA)/Ro antibodies (Ab), anti-SSB/La Ab, anti-nuclear Ab (ANA) and rheumatoid factor (RF) IgM in 113 patients with primary Sjögren's syndrome (pSS).

A. Ab to SSA, ANA and RF IgM co-existed in 40.7% of the patients (grey) and 31.0% of the patients were positive for both anti-SSA and anti-SSB Ab (dotted line). Only 4 patients were ANA and RF positive but anti-SSA Ab negative.

B. The presence of the triple association (ANA+SSA+RF) resulted in more severe disease with salivary gland (SG) involvement, and abnormal SG ultrasonography (SGUS).

Fisher's exact test was used to measure the odds ratio and the corresponding 95% confidence interval. Associations are indicated when  $p<0.05$ .



nescence immunoanalyzer (ISYS-IDS; Immunodiagnostic system Ltd, Bordon, UK) and consumables from Technogenetics (Milan, Italy) which were selected to assay 17 autoantigens including centromere (Cent-B), dsDNA, Sm, U1-snRNP, Scl70, Jo1, cardiolipin (IgG & IgM),  $\beta_2$ -GPI (IgG & IgM), CCP2, PR3, MPO, GBM, transglutaminase (IgA), and deamidated gliadin (IgA & IgG). Atypical Abs in current practice were detected in 1/15 (6.7%) patient from group I and in 2/29 (6.7%) patients from group II. These atypical Ab were directed against Cent-B, Jo1, and dsDNA plus IgG deamidated gliadin Ab, but without associated clinical signs.

In conclusion, our results testing typical and atypical Ab in pSS showed (i) that ANA, anti-SSA/Ro Ab, anti-SSB/La Ab, and RF are strongly associated; (ii) that patients with more severe disease, abnormal SG ultrasonography and lymphocyte activation are positive for ANA+SSA+RF; (iii) that combining anti-SSA/Ro Ab detection with ANA and RF, rather than with anti-SSB/La Ab, improves the characterisation of patients with an active immunological profile; and (iv) that unconventional Ab are exceptional and detected in a minor fraction of patients.

C. CAPALDO<sup>1</sup>  
G. CARVAJAL ALEGRIA<sup>1,2</sup>  
D. CORNEC<sup>2,3</sup>  
S. JOUSSE-JOULIN<sup>2,3</sup>  
V. DEVAUCHELLE-PENSEC<sup>2,3</sup>  
Y. RENAUDINEAU<sup>1,3</sup>

<sup>1</sup>Laboratory of Immunology and Immunotherapy, CHU Morvan, Brest, France;  
<sup>2</sup>Unit of Rheumatology, CHRU Cavale Blanche, Brest, France; <sup>3</sup>INSERM ESPRI, ERI29/EA2216, Innovative Medicines Initiative PRECISEADS, European University of Brittany, Brest, France.

Address correspondence to:

Prof. Yves Renaudineau, Laboratory of Immunology and Immunotherapy, Brest University Medical School Hospital, BP824, F29609, Brest, France.

E-mail: yves.renaudineau@univ-brest.fr

Competing interests: none declared

## References

- VITALI C, BOMBARDIERI S, JONSSON R *et al.*: Classification criteria for Sjögren's syndrome: a revised version of the European criteria proposed by the American-European Consensus Group. *Ann Rheum Dis* 2002; 61: 554-8.
- SHIBOSKI SC, SHIBOSKI CH, CRISWELL L *et al.*: American College of Rheumatology classification criteria for Sjögren's syndrome: a data-driven, expert consensus approach in the Sjögren's International Collaborative Clinical Alliance cohort. *Arthritis Care Res* (Hoboken) 2012; 64: 475-87.
- LUCIANO N, VALENTINI V, CALABRO A *et al.*: One year in review 2015: Sjögren's syndrome. *Clin Exp Rheumatol* 2015; 33: 259-71.
- CORNEC D, SARAUX A, JOUSSE-JOULIN S *et al.*: The Differential Diagnosis of Dry Eyes, Dry Mouth, and Parotidomegaly: A Comprehensive Review. *Clin Rev Allergy Immunol* 2015; 49: 278-87.
- BOURNIA VK, VLACHOYIANNPOULOS PG: Subgroups of Sjögren syndrome patients according to serological profiles. *J Autoimmun* 2012; 39: 15-26.
- BAER AN, MCADAMS DEMARCO M, SHIBOSKI SC *et al.*: The SSB-positive/SSA-negative antibody profile is not associated with key phenotypic features of Sjögren's syndrome. *Ann Rheum Dis* 2015; 74: 1557-61.
- RAMOS-CASALS M, SOLANS R, ROSAS J *et al.*: Primary Sjögren syndrome in Spain: clinical and immunologic expression in 1010 patients. *Medicine* (Baltimore) 2008; 87: 210-9.
- TER BORG EJ, RISSELADE AP, KELDER JC: Relation of systemic autoantibodies to the number of extraglandular manifestations in primary Sjögren's Syndrome: a retrospective analysis of 65 patients in the Netherlands. *Semin Arthritis Rheum* 2011; 40: 547-51.
- THEANDER E, MANDL T: Primary Sjögren's syndrome: diagnostic and prognostic value of salivary gland ultrasonography using a simplified scoring system. *Arthritis Care Res* (Hoboken) 2014; 66: 1102-7.
- FAUCHAIS AL, MARTEL C, GONDRA N *et al.*: Immunological profile in primary Sjögren syndrome: clinical significance, prognosis and long-term evolution to other auto-immune disease. *Autoimmun Rev* 2010; 9: 595-9.