ILC3) via increase of Fas (CD95) expression, rendering the ILC more susceptible to apoptosis, Maazi JACI 2017, Zhang JCI 2015. Duerr Nat Immunol 2016

Objectives: In this study, we explored for the first time the frequency and phenotype of circulating ILCs in pSS and SLE and their relation to the IFN signature.

Methods: Frequencies and phenotypes of ILC subsets and pDCs were assessed by flow cytometry in peripheral blood of patients with pSS (n=20), SLE (n=20) and healthy controls (n=17). Patients were stratified by the presence or absence of an IFN signature as assessed by RT-qPCR on peripheral blood mononuclear cells as previously described. Brkic ARD 2013

Results: Results ILC1 numbers were increased in peripheral blood of patients with SLE as compared to healthy controls and in pSS patients ILC1 numbers correlated with disease activity (ESSDAI score), serum IgG levels and anti-SSB auto-antibodies (all $p < 0.05$). Numbers of ILC1, ILC2 or ILC3 did not significantly differ between patients with SLE and pSS. However, patients with a high expression of the type I IFN signature had significantly decreased numbers of ILC2 and ILC3 ($p = 0.04$ and $p = 0.02$, respectively). The decrease of ILC2s and ILC3s was related to increased expression of Fas (CD95) on these cells in patients with a high type I IFN signature (both $p = 0.01$).

Conclusions: Both in SLE and pSS, a type I IFN signature is related to reduced numbers of circulating ILC2s and ILC3s in association with increased Fas expression on these cells possibly rendering them more susceptible to Fas/FasL-dependent apoptosis at peripheral sites.

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FRI0294

INJURED PODOCYTES EXPRESS COMPLEMENT FACTOR H AND PROCESS IMMUNE COMPLEX DEPOSITION IN GLOMERULAR SUBENDOTHELIAL AREA

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Background: Glomerular Immune complex (IC) deposition causes glomerulonephritis in lupus nephritis. The reaction of intrinsic glomerular cells differs according to the area of IC deposition. Subepithelial IC deposits cause functional and structural changes on podocytes (glomerular visceral epithelial cells) through complement pathway activation as is well known in membranous glomerulonephritis. In this setting, podocytes can induce some complement factors including complement factor H (CFH) which serves not only to regulate the alternative pathway but also to process subepithelial IC deposition.¹ However, whether podocyte processing of IC deposition is limited to only the subendothelial area is obscure.

Objectives: To clarify the role of podocytes in IC deposition in the glomerular sub-endothelial area.

Methods: NEP25 mice genetically expressing human CD25 in podocytes were used. Intravenous immunotoxin for human CD25 (LMB2) provokes podocyte-specific injury (NEP25/LMB2). By shortening the period of LMB2 exposure to NEP25 mice, we mitigated the podocyte injury. We administered intraperitoneally IgG3-producing hybridoma clones, 2B11.3, which were previously established from an unmanipulated MRL/lpr mouse, to NEP25 mice (NEP25/hybridoma). Furthermore, we also injected short-term LMB2 to NEP25/hybridoma mice (NEP25/hybridoma/LMB2). We investigated IC deposits by immunofluorescence and electronic microscopic study. We measured complement regulatory factor mRNA expression including CFH, complement factor I (CFI), decay-accelerating factor (DAF), complement receptor 1-related gene/protein y (Cry), C3a receptor (C3aR) and C5a receptor (C5aR) of isolated glomeruli of each mouse by quantitative real time-PCR. In an *in vitro* study, we assessed CFH mRNA expression of immortalised mouse podocytes injured by puromycin.

Results: First, NEP25/LMB2 mice showed glomerular tuft collapse with epithelial cell hyperplasia, suggesting podocyte loss by light microscopy study as reported previously. mRNA expression of all complement regulatory factors but C5aR was decreased in NEP25/LMB2 mice as compared with NEP25 mice. This result suggests that podocytes produce these complement regulatory factors. On the other hand, when the podocyte injury was mitigated, CFH and C3aR mRNA expression increased, and CFI, DAF and Cry mRNA expression decreased as compared with NEP25 mice. Second, we detected IC deposition only in the glomerular sub-endothelial area without endocapillary proliferative lesion in NEP25/hybridoma mice. They showed increase of C3aR mRNA expression, and decrease of CFI and DAF mRNA expression as compared with controls. Notably, subendothelial IC deposition did not alter CFH mRNA expression. Third, NEP25/hybridoma/LMB2 (milder podocyte injury) showed decrease of IC deposits in glomeruli and increase of only CFH mRNA expression (1.7-fold) as compared to NEP25/hybridoma mice, while there was no significant change in mRNA expression of other

complement regulatory factors. Finally, puromycin-induced mild podocyte injury promoted CFH mRNA expression in cultured podocytes as compared to controls.

Conclusions: Mildly injured podocytes induce CFH expression, and serve to process IC deposition in the glomerular subendothelial area.

REFERENCE:

[1] J Am Soc Nephrol 2007;18:1157–66.

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FRI0295

INHIBITION OF CATHEPSIN S LEADS TO SUPPRESSION OF ANTIGEN SPECIFIC T CELLS FROM PATIENTS WITH PRIMARY SJÖGREN SYNDROME

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Background: Primary Sjögren syndrome (pSS) is an autoimmune disease characterised by an infiltration of T and B cells into exocrine gland tissue and its subsequent destruction¹. Antigen presenting cells, including B cells, foster T cell activation and anti-SS-A/SS-B producing plasma cells, eventually leading to disease progression and systemic complications².

Objectives: Cathepsin S (CatS) is crucially involved in MHCII processing in pSS mouse models³ and patients⁴. In this translational study we investigated the *ex vivo* effects of the CatS inhibitor RO5459072 in different bio-compartments, including specific T cells, of pSS patients and healthy controls.

Methods: *Ex vivo* CatS activity was assessed in different bio-compartments of 15 pSS patients and 13 healthy controls and in presence or absence of RO5459072 using commercial activity and quantification assays. In addition, antigen (5 µg/mL SS-A, 5 µg/mL SS-B, 5 µg/mL Influenza H₃N₂; 2 µg/mL Tetanus Toxoid and 100 ng/mL SEB) specific T cell responses were examined using 2 × 10⁵ PBMC/well IFN-γ/IL-17 Dual ELISPOT (48 hour incubation) and 5 × 10⁴ PBMC/well BrdU proliferation assays after (72 hour incubation) in presence or absence of RO5459072.

Results: pSS patients showed significantly higher CatS activity in tear fluid than healthy controls (two-tailed t-test $p < 0.01$). RO5459072 significantly suppressed CatS activity in tears of pSS patients (two-tailed t-test $p < 0.01$). CatS inhibition also exerted a strong and dose-dependent suppression of T cell responses towards SS-A and SS-B antigen in *ex vivo* derived pSS patient cells in Elispot and BrdU assays (table 1).

Table 1 Suppression of antigen specific T cell responses (Elispot) and suppression of cell proliferation (BrdU) in the absence or presence of RO5459072 (0–100 µM). One-tailed t-test: * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$, **** $p < 0.0001$

Conclusions: CatS activity in tear fluid seems to be a relevant biomarker for pSS disease activity. RO5459072 is a potent inhibitor of CatS and the pSS associated relevant antigen specific T cell responses.

REFERENCES:

- [1] Voulgarelis M, Tzioufas AG. Pathogenetic mechanisms in the initiation and perpetuation of Sjogren's syndrome. Nat. Rev. 2010;6:529–537.
- [2] Brito-Zeron P, Baldini C, Bootsma H, et al. Sjogren syndrome. Nat Rev Dis Primers 2016;2:16047.
- [3] Saegusa K, Ishimaru N, Yanagi K, et al. Cathepsin S inhibitor prevents autoantigen presentation and autoimmunity. J Clin Invest 2002;110(3):361–9.
- [4] Hamm-Alvarez SF, Janga SR, Edman MC, et al. Tear cathepsin S as a candidate biomarker for Sjogren's syndrome. Arthritis Rheumatol 2014;66(7):1872–81.

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