Background: Renal fibrosis is a feared complication of Lupus Nephritis (LN), and is associated with irreversible loss of kidney function. In our previous experiments, we found that intrarenal infiltration by immune effectors in LN correlates with the development of renal fibrosis. Here, we wondered whether cellular senescence, through its typical secretome (known as senescence-associated secretory phenotype or SASP) or through the accumulation of functionally incompetent cells, are part of the renal functional impairment and fibrotic process in LN.

Methods: Microarray data (Illumina HumanHT-2 v4 Expression BeadChip), obtained by our group from 32 human LN kidney biopsies and 8 controls were mined using GeneSpring software in order to study the expression of SASP-associated transcripts. Senescent cells were identified in human LN kidney biopsies using an anti-p16 antibody (Roche Diagnostics). Evaluation of glomerular activity and chronicity indices, glomerular and interstitial fibrosis was performed using conventional or quantitative scores on HE-, PAS-, and Red Sirius-stained sections. Clinical and biological data were retrieved from the medical files of the patients.

Results: Mining of microarray data obtained from 32 LN kidney biopsies indicated that SASP-associated transcripts (e.g., IGFB4, VCAM1, TGFβ2, COL1A2, MMP7) were significantly overexpressed in kidney biopsies characterized by the presence of adaptive immune cell infiltrates in the interstitium or through the accumulation of functionally incompetent cells. Expression of SASP-associated transcripts correlated significantly with the expression of β galactosidase of the secretory phenotype (GLB1), a key regulator of the senescence process.

Conclusions: In a pilot experiment, we stained LN renal biopsy sections using anti-p16 antibodies, in order to detect the presence of senescent cells. We found a positive stain in podocytes and renal tubular cells from 8 LN patients. The number of positive cells correlated positively with the number of intrarenal CD8-positive cells and the amount of fibrosis in these samples, while it correlated negatively with renal function (eGFR).

Conclusion: Our data show that the presence of senescent podocytes and renal tubular cells in LN kidney biopsies correlates with fibrotic changes and impaired renal function. Characterization of senescent cells in a larger cohort of LN biopsies is ongoing. Our observations are in line with the hypothesis that inflammation-accelerated senescence links the presence of activated adaptive immune effectors in the lupus kidney and the development of fibrosis.
Acknowledgements ERC (No. 742390).

REFERENCE


P104 CORRELATION BETWEEN INTERSTITIAL CD8+ T CELL INFILTRATION AND FIBROTIC PROCESSES IN A MOUSE MODEL OF LUPUS NEPHRITIS

1,2Pauline Montigny, 1Àurelie Degroof, 3Davide Busa, 1,3Frédéric Houssiau, 1Bernard Lauwerys. 1Pôle de Pathologies Rhumatologiques Systémiques et Inflammatoires, UCLouvain, Louvain; 2Service de Rhumatologie, CHU UCL Namur, Yvoir; 3Plateforme de Cytométrie de Flux, Institut de Recherche Expérimentale et Clinique, UCLouvain, Louvain; 4Service de Rhumatologie, Cliniques Universitaires Saint Luc, Bruxelles, Belgium

Background Tubulo-interstitial damage during lupus nephritis (LN) is associated with poor renal prognosis in the long run. Here, we describe the progression of tubulo-interstitial fibrosis and immune cells infiltration with emphasis on CD8+ T cells, in parallel with renal outcomes in a mouse lupus model.

Methods We collected blood, urine and kidneys from 34 B6/Sle1.Sle2.Sle3 lupus-prone mice, before disease onset and at different stages of disease progression. RNA was extracted from kidneys and hybridized on Mouse Gene 2.0 ST exon arrays. Histopathological scores (NIH Activity and Chronicity Indexes) and digital quantification of fibrosis, IgGK deposits, CD8+ and CD3+ T cells were performed on total kidney.

Indexes) and digital quantification of fibrosis, IgGK deposits, CD8+ and CD3+ T cells were performed on total kidney. Renal CD8+ T cell phenotypes were determined by flow cytometry. Plasma urea and albuminuria were measured by immunoenzymatic assays.

Results IgGK deposits, CD8+ T cell infiltration and interstitial fibrosis increase with the progression of renal disease, evaluated by histopathological scores and plasma urea. Further, digital quantifications allowed us to identify a significant correlation (r=0.45, p=0.011) between local CD8+ T cell population and fibrosis, while total CD3+ cells population and IgGK deposits did not display such association. Moreover, characterization of CD8+ T cell subpopulations showed that fibrosis is more specifically linked to effector functions of CD8+ T cells. Transcriptomic analyses supported this association, with a high correlation coefficient between mean expression of effector functions transcripts and the presence of a fibrotic signature (r=0.92, p<0.0001).

Conclusions Our results support the association between CD8+ T cell tubulo-interstitial infiltration and renal outcomes in a mouse lupus model. Further, a strong correlation is identified between effector functions of CD8+ T cells and fibrotic processes, opening new avenues of research in the pathogenesis of LN.

Acknowledgements This work was supported by F.R.S.-F.N.R.S and by Fondation Roi Baudouin.

P105 EXPRESSION DIVERSITY OF INTERFERON-STIMULATED GENES IN PERIPHERAL BLOOD CELLS FROM PATIENTS WITH SYSTEMIC LUPUS ERYTHEMATOSUS

Kanwal Siddiqi, Søren Jacobsen. Copenhagen Lupus and Vasculitis Clinic, Rigshospitalet, Copenhagen University Hospital, Copenhagen, Denmark

Background A central facet of the immunopathogenesis of systemic lupus erythematosus (SLE) is the activation of interferon (IFN) signalling. In SLE patients, several types of IFNs are upregulated as well as the genes that are stimulated by IFNs. Sustained expression of interferon-stimulated genes (ISGs) may have adverse effects including altered T cell function, tissue inflammation and organ damage. The distinct association of the IFN gene signature to SLE disease activity is still uncertain.

Methods Peripheral blood samples from 34 SLE patients and 15 healthy controls were collected in PAXgene tubes. RNA from peripheral blood cells (PBCs) was extracted using the PAXgene Blood RNA Kit. The mRNA transcripts of 105 ISGs were measured using the multiplexed Nanostring nCounter Gene Expression platform. Bioinformatics and statistical analysis were performed using nSolver and SPSS.

Results We found that SLE patients had significantly higher expression levels of a wide range of ISGs, as compared to healthy controls. The 5 most upregulated ISGs in PBCs were IFI27, IFI44L, USP18, RSAD2 and ISG15. Using principal component analysis we identified two subsets of ISGs of which one included the mentioned top-five expressed ISGs; this gene cluster consisted of ISGs with predominantly antiviral functions and STAT1/STAT2, and did not associate with clinical disease activity or history of nephritis in SLE patients. Instead these genes associated with anti-SSA/SSB antibodies and PBC expression of PDCD1 and LAG3, which are markers of CD8 T cell exhaustion. The second group of ISGs associated with clinical disease activity, STAT3 and NFkB expression.

Conclusions Our results indicate the presence of ISG subsets that are differentially associated to clinical, serological and immunological features of SLE. Thus, the IFN gene signature in SLE may consist of a highly complex network of ISGs that can be clustered, and each in their way contribute to the pathogenesis of SLE. Also, this challenges the construction of IFN gene signatures and scores with respect to what purpose they are intended for.

P106 PENTAMERIC, BUT NOT MONOMERIC C-REACTIVE PROTEIN, LIMITS THE SNRNP-IMMUNE COMPLEX TRIGGERED TYPE I INTERFERON RESPONSE: IMPLICATIONS FOR LUPUS PATHOGENESIS

1Cecilia Svanberg, 1Helena Enocsson, 2Klara Martinsson, 2Lawrence Potempa, 1Ibraheem Rajabi, 1Jonas Wetterö, 1Marie Larsson, 1Christopher Sjöwall, 1Linköping University, Linköping, Sweden; 2Roosevelt University, Chicago, USA

Background Systemic Lupus Erythematosus (SLE) is an autoimmune systemic disease affecting multiple organs and which is characterized by autoantibodies directed against nuclear constituents. Common autoantibody targets include double-stranded (ds) DNA and small nuclear ribonucleoproteins (snRNPs; i.e. U1-snRNP). Uptake of immune complexes (ICs) by plasmacytoid dendritic cells (pDCs) can activate endosomal toll-like receptors (TLRs) such as TLR-7 and TLR-9 if nucleic acids are present in the ICs. Such activation is dependent on IC internalization by Fcy receptor type IIa (FcyRIIa), and results in production of type I interferons (IFNs), a hallmark of SLE and a target for therapeutic interventions. The acute phase protein C-reactive protein (CRP)