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Adaptive immunity (T cells and B cells) in rheumatic diseases

FOSL-2 IS A REPRESSOR OF FOXP3 EXPRESSION DURING TREG DEVELOPMENT AND CONTROLS AUTOIMMUNITY

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Background: Fos like 2 (Fosl-2) is a transcription factor belonging to the AP-1 transcription complex. We have recently described a Fosl-2 transgenic (tg) mouse model which develops a multi-organ inflammatory and autoimmune phenotype. In these mice, we have characterized a decrease in regulatory T cells (Tregs), which preceded the activation of T cells and the development of inflammation.

Objectives: To analyze how Fosl-2 reduces the Treg population and triggers autoimmunity in Fosl-2 tg mice.

Methods: We used previously generated Fosl-2 tg overexpressing and Fosl-2 T cell specific knock out (Fosl-2 ko) mice. For mixed bone marrow reconstitution experiments, lethally irradiated recipients received a one to one mix of Fosl-2 tg and wt CD4 T cells after 24 hours of stimulation with anti-CD3 (2ug/ml) and anti-CD28 (2ug/ml).

Results: We first addressed whether Fosl-2 affected Tregs in a cell intrinsic way using mixed bone marrow experiments. In these animals, the CD45.2 Fosl-2 tg CD4 T cells showed a much lower proportion of Tregs compared to the CD45.1 wt population, both in the spleen (0.73±0.15 vs 31.6%±3.6, P=0.002) and thymus (1.3%±0.15 vs 3.23%±0.78, P=0.001). This demonstrates that Fosl-2 overexpression represses Treg development in a cell intrinsic way.

In T cell transfer experiments, Rag2+ mice receiving 10^5 Fosl-2 tg CD4 T cells developed lung inflammation 5 weeks after transfer, confirming that T cells are inducers of inflammation in Fosl-2 tg mice. Moreover, co-transfer of either 3*10^5 or 10^6 wt Treg cells resulted in a dose dependent reduction of inflammation. These data indicated that the decrease in the Treg population in Fosl-2 tg mice is responsible for the induction of inflammation.

We then analysed Fosl-2 transcriptional targets in T cells by RNA-seq. Using a fold change > 1.5 and False Discovery Rate (FDR) of 0.05, we identified 191 differentially expressed genes in both Fosl-2 tg and Fosl-2 ko compared to wt. Interestingly, one of the top target genes of Fosl-2 was FoxP3. This unbiased approach thus revealed that FoxP3 expression is repressed by Fosl-2, with a 6.5 fold reduction in Fosl-2 tg cells and a 2.5 fold increase in Fosl-2 ko cells. This effect was confirmed on the protein level with a reduction in FoxP3 induction in Fosl-2 tg cells treated with TGFβ. The repression of Treg development observed in Fosl-2 tg mice could thus be explained by a direct transcriptional control of FoxP3 expression. Additionally, we found that Fosl-2 repressed a set of genes important for Tregs and other T helper cells. This included Nlrp2, a transcription factor involved in Treg development, IRF8 and Eomes, two genes involved in the polarization of Th1 and Th17 cells, or Ccl1, a chemokine important for Treg homeostasis.

Conclusion: Fosl-2 is involved in the control of FoxP3 expression in T cells. Through this, overexpression of Fosl-2 represses Treg development and induces a Treg dependent autoimmune phenotype in mice. This mechanism could thus be involved in the pathogenesis of autoimmune diseases and might represent a therapeutic target to modulate the Treg population.

Disclosure of Interests: Florian Renoux: None declared, Mara Stellato: None declared, Alexander Vogelseder: None declared, Riyung Huang: None declared, Anun Subramaniam: None declared, Przemyslaw Blyszczuk: None declared, Jörg Distler: None declared, Gabriela Kania: None declared, Onur Boyman: None declared, Oliver Distler: Grant/research support from: Prof. Distler received research funding from Actelion, Bayer, Boehringer Ingelheim and Mitsubishi Tanabe to investigate potential treatments of sclerodermia and its complications, Consultant for: Prof. Distler had/had consultancy relationship within the last 3 years with Actelion, AmNar, Bayer, Boehringer Ingelheim, ChemomAb, espereFoundation, Genentech/Roche, GSK, Inventiva, Italfarmaco, IQvia, Lilly, medac, MedImmune, Mitsubishi Tanabe Pharma, Pharmaciesci, Novartis, Pfizer, Sanofi, Serodapharm and UCB in the area of potential treatments of sclerodermia and its complications. In addition, he had/had consultancy relationship within the last 3 years with A. Menanini, Amgen, Abbvie, GSK, Medpharma, MSD, Pfizer and UCB in the field of arthritis and related disorders


DIFFERENTIAL RECONSTITUTION OF B-CELL SUBSETS IN SYSTEMIC SCLEROSIS PATIENTS AFTER AUTOLOGOUS HEMATOPOIETIC STEM CELL TRANSPLANTATION

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Background: Autologous hematopoietic stem cell transplantation (AH SCT) is nowadays a clinical reality as an effective and safe approach to treat systemic sclerosis patients’ refractory to conventional therapy using immune suppressive drugs, although its immune mechanisms are still not completely understood.

Objectives: To evaluate the reconstitution of B-cell subsets in SSc patients following AHSC.

Methods: Peripheral blood samples were harvested from twenty-four SSc patients before transplantation and at 30, 60, 120 and 180 and 360 days post-AHSC. The immunophenotyping, regulatory B-cell IL-10 production and suppressive assays were assessed by flow cytometry.

Results: Compared to baseline, naïve B-cells (CD19+CD27+IgD-) significantly decreased in frequency and absolute counts at 30 days post-AHSC, followed by an increase at 360 days. There was a transient decrease of non-class-switched memory-B-cell (CD19+CD27+IgD+) frequency at 30 days, followed by an increase at 360 days. In addition, mature class-switched memory-B-cells (CD19+CD27+IgD-) and plasma cell (CD19+CD27+IgM+IgD+) frequency at 30 days post-AHSC, while the frequency of double-negative B-cell (CD19+CD27+IgD+) increased at 30 days post-AHSC.

Conclusion: Following transplantation, SSc patients displayed increased naïve B-cells values and decreased memory B-cells, which might contribute to self-tolerance reestablishment, disease remission and clinical improvement.

REFERENCES