CD4/CD8 CD28 null cells and HCMV in the pathogenesis of Systemic Sclerosis (SSc).

Gnaneshwer J.[1], Bason C.[1], Argentino G.[1], Beri R.[1], Tinazzi E[1], Puccetti A.[2], Lunardi C[1].

[1] Department of Medicine, University of Verona, Italy.

[2] University of Genoa, Genoa, Italy.

Background: Systemic sclerosis (SSc) is a chronic inflammatory connective tissue disease, characterized by vascular dysfunction, immune alteration, and tissue fibrosis. Several studies involving both humans and animal models point towards a role for T cells in the pathogenesis of SSc, however, the precise phenotype, function, and specificity of pathogenic T cells remain elusive. Moreover, several studies reported increased frequencies of a T cell subset, known as CD4+/CD8+CD28null T cells, in peripheral blood in various autoimmune diseases or chronic inflammatory disorders or infectious diseases such as HCMV infection. Such CD4+/CD8+CD28null T cells are highly differentiated T cells lacking the co-stimulatory molecule CD28; such cells acquire expression of other receptors commonly associated with natural killer cells and display proinflammatory, cytotoxic and apoptosis-resistant features. In contrast to CD28null T cells, regulatory T cell subset is critical for maintaining immune tolerance and has also been described to assist in the tissue repair process. The aims of my project are to investigate CD4+/CD8+CD28null and CD57+CD4+/CD8+CD28null T cell subsets in peripheral blood of patients with SSc, by evaluating frequencies, phenotype, function and clinical relevance of these cells. Along with CD28null T cells, regulatory T cells were also investigated. Furthermore, we checked whether Iloprost (a Prostacyclin analog, commonly used in SSc treatment) may have any effect in the modulation of the percentage of these T cell subsets. We correlated these T cell subsets with disease subtype and patients'age. Finally, we have analyzed whether the percentage of these T cell subsets correlates with anti-HCMV antibodies.

Materials and Methods: We studied 70 SSc patients and 30 healthy donors blood samples in order to evaluate the percentage of the different T cell subsets by FACS analysis, using the following antibodies CD4 (APC-H7) CD8 (PE and FITC) CD25 (FITC), CD28 (PC7) CD57 (PE), and CD127 (PC7). We have also analyzed the possible variation of the percentage of these T cell subsets after treatment with prostacyclin analog and the correlation we organ involvement and with anti-HCMV antibodies.

Results: an increased number of CD4+/CD8+CD28nullCD57+ T cells in SSc patients compared to HD was found. Defects in both subsets of Treg cells and fluctuations have been described in several autoimmune diseases compared to healthy controls. However, frequencies of both CD4 and CD8 Treg cells in patients and healthy controls were similar. T cell subsets frequency were studied in SSc patients divided in different age group and only CD4+CD28nullCD57+ T cells were positively correlated with age (P=0.0124). No correlation was found in the CD4/CD8/CD28null T cells before and after Iloprost infusion both for one day and for 5 consecutive days. We then compared the different T cell subsets frequency in SSc patients with and without organ involvement. CD8+ (P=0.0358), CD4+CD28null (P=0.0150), CD4+CD28nullCD57+, CD4 Treg (P=0.0133)and CD8 Treg (P=0.0028) cells showed a statistically significant difference according to organ involvement. Moreover CD28 null cells correlate with the presence of anti-HCMV antibodies in SSc patients.

Conclusion: The increased number of CD4+/CD8+CD28nullCD57+ T cells in SSc patients compared to healthy donors and their correlation with the presence of anti-HCMV antibodies further support the hypothesis of an involvement of HCMV in the pathogenesis of SSc as we have previously reported.