Title

Negative Immune Checkpoint Molecules on T Regulatory Cells Distinguish RA, SLE and Healthy Controls

Authors and Affiliations

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Background/Purpose

T regulatory cells (Tregs) play a crucial role in the regulation of the immune response and are of utmost interest when studying autoimmune diseases, such as rheumatoid arthritis (RA) and systemic lupus erythematosus (SLE). Therefore, targeting specific Treg markers in therapy is now widely discussed. Expression of T cell immunoglobulin 3 (TIM-3) is associated with an enhanced immune suppression and are currently discussed as target in cancer therapy [1]. Fc receptor-like protein 3 (FCLR-3), which regulates Treg proliferation, was shown to be associated with the susceptibility in juvenile RA [2]. Other surface markers, like CD161 enable Tregs to produce IL-17A, IFNg and IL-2, hence promoting inflammation [3]. The aim of this study was to distinguish between RA and SLE using anti-and pro-inflammatory cell markers on Treg subsets.

Material and Methods

Peripheral blood samples from 66 RA patients (mean ± SD; age 60 ± 10 years, female ratio: 0.68, disease duration 18 ± 14 years), 40 SLE patients (age 42 ± 13 years, female ratio 0.85, disease duration 11 ± 13 years) and 72 age-matched healthy participants (age 46 ± 17 years, female ratio 0.68) were drawn over a sampling period of 2 years. Freshly isolated PBMCs were stained and Treg subsets were identified by the expression of CD25, CD127, FoxP3, CD45RA and CD15 on the surface of CD3 and CD4 positive T cells. $CD25^{+}CD127^{+}CD45^{-}$ Tregs were further subclassified by the expression of TIM-3 (CD366) and FCLR-3 (CD307c). CD161 was used to identify Th17 type Tregs ($CD15S^{-}CD161^{+}$) and transitional Tregs ($CD15S^{-}CD161^{-}$). All cytometric measurements were performed using a standardized BD LSRFortessa platform.

Results

Transitional Tregs ($CD15S^{-}CD161^{-}$) were significantly higher ($p < 0.001$) in RA patients compared to the SLE and healthy cohort (40.5 ± 13.4% vs. 28.7 ± 9.6% and 29.7 ± 9.4% respectively). However, differences in the $CD161^{+}$Th17 type Treg population could not be detected. Tregs expressing TIM-3 were higher in both RA and SLE patients compared to healthy controls (2.8 ± 2.3%, $p = 0.0105$ and 2.6 ± 1.6%, $p = 0.0031$ vs. 0.8 ± 0.7% respectively), but did not differ between the rheumatic diseases. On the other hand, FCLR-3^{+}Tregs distinguished RA and SLE patients (17.8 ± 13.3 vs. 25.3 ± 13.1%, $p = 0.0036$), as well as SLE patients and healthy controls (16.8 ± 12.9%, $p = 0.0112$). No findings were correlated with the disease activity of RA or SLE patients.

Conclusion

Expression of negative immune checkpoints TIM-3 and FCRL-3 on Tregs not only distinguish healthy controls from RA and SLE patients but can be used to differentiate between different rheumatic diseases. These findings indicate that Tregs trigger the regulation of the immune response in RA and SLE, yet the activation of different Treg subsets is disease-specific.

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