IJIR. 2018;2(1):A5 DO:10.15305/ijir/v2i1/284

ORAL ABSTRACTS

Molecular interaction of T cell receptor and chemokine receptor CCR6 signaling in the differentiation of pathogenic Th17 cells in autoimmunity

Meitei HT[1]*, Shirolkar A[2], Rapole S[3], Lal G[4]

[1-4]National Centre for Cell Science, Pune, India

CCR6 is known to express on inflammatory Th17 and regulatory CD4 T cells (Tregs), and drives the migration into the CCL20 rich inflamed tissues. Recently, we showed that CCL20 promotes differentiation of Th17 cells as well as pathogenic Th1-like Th17 cells, and inhibit the function of Tregs. Interaction of T cell receptor (TCR) and CCR6 signaling in the differentiation and function of Th17 and Treg cells is not well characterized. To investigate the TCR signaling and CCR6 signaling, we isolated CD4+CD25-CD44- (Naïve) CCR6gfp+ and CCR6gfp-T cells from CCR6gfp/- mice using flow cytometry sorting. Naïve CD4 T cells were *in vitro* differentiated into Th17 and Treg cells in presence or absence of purified recombinant CCL20 and anti-CD3e antibody, and expression and phosphorylation of various signaling molecules were analyzed using flow cytometry and Western blot, and differential protein expression in these cells were analyzed using mass spectrometry based label-free quantitative proteomics approach.

Our results showed that CCL20 promoted the phosphorylation of TCR signaling molecules such as LCK and SRC in a dose- and time-dependent manner. Furthermore, CCL20 stimulation increased the phosphorylation of ZAP70 and induced the co-localized of ZAP70 with CD3ζ in T cells. Our data showed that CCL20 also inhibited the phosphatase PTPN22 expression in a time-dependent manner, and modulated the TCR signaling. Purified Th17 cells from colitic mice also showed increased phosphorylation of PI3 kinase and lower PTPN22 expression as compared to Foxp3+Treg cells. Mass-spectrometry based quantitative proteomic analysis showed that CCL20 changed the expression of several important proteins during CD4 T cells differentiation. Together, these data suggest that CCR6 signaling interact with TCR signaling during inflammatory condition and drive the differentiation of Th17 cells.

Presented on: 19 August 2018
Presented by: Meitei HT