Understanding aberrant IL-6 pre-exposure of CD4+ T-cells in early rheumatoid arthritis.

Laura Ridgley[1, 2], Amy Anderson[1, 2], Andrew Skelton[2, 3], David Young[2], John Isaacs[1, 2], Ruaidhri Carmody[1, 3], Arthur Pratt[1, 2]

[1]Arthritis Research UK, Rheumatoid Arthritis Pathogenesis Centre of Excellence (RACE)
[2]Musculoskeletal Research Group, Institute of Cellular Medicine, Newcastle University, Newcastle upon Tyne, UK
[3] Bioinformatics Support Unit, Faculty of Medical Sciences, Newcastle University, Newcastle upon Tyne, UK
[4] Institute of Infection, Immunity and Inflammation, University of Glasgow, Glasgow, UK

Background

A -12-gene signature in CD4+ T-cells had discriminatory utility for early rheumatoid arthritis (RA) patients compared with disease controls. This signature is enriched for STAT-3 target genes whose expression correlates with paired circulating IL-6. I hypothesise that pre-exposure of CD4+ T-cells to IL-6 mediates STAT-3 activation and aberrant effector function following T-cell receptor (TCR) stimulation, providing a mechanism of antigen non-specific immune dysfunction in early RA.

Methods

A model for cytokine pre-exposure was developed, in which naïve (CD45RA+) and antigen experienced (CD45RA-) CD4+ T-cells from healthy human donors were cultured with IL-6 and equimolar soluble IL-6R for 3 days, before being washed and stimulated with anti-CD3/anti-CD28 for 6 days. RNA was extracted at multiple experimental time-points and global gene expression profiling undertaken. Phenotype and proliferation were assessed by flow cytometry, measuring cell surface markers and CFSE dilution. Whether the observed consequences of IL-6 pre-exposure reflected transcriptional and phenotypic characteristics of CD4+ T-cells isolated from patients with early RA was explored.

Results

The effects of IL-6 pre-exposure were seen most prominently in naïve CD4+ T-cells, potentially related to IL-6 receptor expression. Pre-exposure of healthy control naïve CD4+ T-cells to physiological levels of IL-6 caused significant STAT-3 gene induction, mirroring genes previously found to distinguish RA patients from disease controls. Following TCR-stimulation, a unique set of genes differentially induced in IL-6 pre-exposed cells were related to cell proliferation and survival. This is consistent with altered effector phenotype of IL-6 pre-exposed cells characterised by dose-dependent enhancement in activation and proliferative capacity as well as altered cytokine profiles. CD4+ T-cells from early RA patients also showed expression of genes differentially induced by IL-6 pre-exposure.

Conclusion

These findings highlight that cytokine “pre-priming” during the early disease state may have consequences for naïve CD4+ T-cell effector function, impacting the transition to disease chronicity in early RA.