DISSECTING THE ROLE OF MICRORNAS IN EFFECTOR *VERSUS* REGULATORY CD4$^+$ T CELL DIFFERENTIATION DURING (AUTO)IMMUNE RESPONSES *IN VIVO*

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Introduction

MicroRNAs (miRNAs) are small non-coding RNAs that negatively regulate gene expression at the post-transcriptional level. They have been implicated in the regulation of the differentiation and function of CD4$^+$ T cell subsets, key players in host defense against pathogens, but also responsible for immune-mediated diseases depending on the correct vs incorrect balance, respectively, between pro-inflammatory effector CD4$^+$T cells, including the IFN-γ-producers T helper 1 (Th)1 and the IL-17-producers Th17 cells, and anti-inflammatory regulatory T cells (Treg). While individual miRNAs were found to regulate the differentiation of specific CD4$^+$ T cell populations, an approach based on *in vivo* responses is still missing and is key to understand how miRNA networks control this balance in pathophysiology.

Methodology

We have established a triple reporter mouse for *Ifng*, *Il17* and *Foxp3,* and subjected it to experimental autoimmune encephalomyelitis (EAE). We performed miRNA-seq analysis on Th1, Th17 and Treg cells isolated from the spleen and lymph nodes (LNs) at peak-plateau stage to identified miRNA candidates specifically expressed in one of the cell populations. We have *in vivo* modulated their expression levels using antagomiRs and observed the course of EAE progression and characterised their upstream regulation *in vitro* in either Th1 or Th17 differentiation conditions.

Results

The miRNA-seq data has allowed the identification of 110 miRNAs differentially expressed between effector (Th1 and Th17) and regulatory (Treg) subsets. From those, 9 were specifically upregulated in one population versus the others. *In vivo* miRNA modulation showed that silencing miR-122 precipitated the onset of EAE, whereas overexpressing miR-1247 decreased the severity of the disease. Cytokine-regulated miR-1247 and miR-122 expression levels inversely associated with pathogenic signatures of Th1 and Th17 cells between lymphoid and central nervous systems.

Discussion

Our results suggest that miR-122 and miR-1247 act as peripheral brakes to $CD4^+$ T cell pathogenicity that are overruled in the inflamed target organ.These findings may have important implications for autoimmune diseases.