A distinct Th17 cell subset produces IL-26 and promotes expression of IL-17 through induction of TGFβ

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Background

IL-26 is a member of the IL-10 cytokine family that is mainly expressed by Th17 cells. IL-26 signals through its heterodimeric receptor IL-10R2/IL-20RA expressed exclusively by epithelial cells but can also activate immune cells in a receptor-independent manner. Indeed, IL-26 is a cationic amphipathic protein with antimicrobial properties that can form complexes with DNA, these complexes can further stimulate immune cells by binding surface proteoglycans leading to the production of inflammatory cytokines (1). Over the past few years, several studies have shown that IL-26 expression is associated with numerous chronic inflammatory diseases like psoriasis, inflammatory bowel diseases or rheumatoid arthritis. However, in contrast to the other Th17 cytokines, little is known on how IL-26 expression is regulated in these cells and how IL-26 is implicated in the development of these diseases. By studying the regulation of IL-26 expression by using psoriasis as a model of Th17-driven inflammation, we show that IL-26-producing Th17 cells are different from IL-17-producing cells and that their generation follows different kinetics and requires a distinct cytokine cocktail. Moreover, we observe that IL-26 can promote the generation of IL-17A-producing Th17 cells through the induction of TGFβ by keratinocytes in a STING dependent manner, demonstrating a potential counter-regulation that may exist *in vivo* between these two Th17 subsets.

Results

1) Dichotomic expression of IL-26 and IL-17A in chronic and acute forms of psoriasis

By analyzing the expression of a panel of cytokines in different forms of psoriasis including acute forms of psoriasis (erythrodermic psoriasis and guttate psoriasis) and chronic plaque psoriasis (**A**), we found a dichotomic expression of IL-26 and IL-17A. IL-26 expression is increased in acute forms of psoriasis whereas IL-17A is highly expressed in chronic plaque psoriasis (**B**). By using confocal microscopy, we found that skin-infiltrating T cells produce high levels of IL-26 in acute forms of psoriasis whereas infiltrating T cells mainly produce IL-17A in chronic plaque psoriasis (**C**).

2) IL-26-producing cells are a distinct Th17 cell subset

By assessing the expression of IL-26 and IL-17A in Th17 cells using a flow FISH technique, we observed that IL-26-expressing cells are different from IL-17A-producing cells (**A**). Moreover, the majority of IL-26+ Th17 cells express only IL-26 and do not co-express the other Th17 cytokines (**B**), indicating that a IL-26 single producers subset exists among Th17 cells.

3) IL-17A- and IL-26-producing Th17 cells have distinct priming requirements

By dissecting the involvement of each cytokine used for the differentiation of Th17 cells, namely IL-1β, IL-6, and IL-23, we found that each cytokine alone was sufficient to induce IL-26 but not IL-17 (**A**). Altogether, the combination of these cytokines increases IL-26 production and the addition of TGFβ to this cocktail is required to induce IL-17A expression (**A**). During the differentiation of Th17 cells, we observed that IL-26 is rapidly expressed as soon as 3 days post-stimulation of naïve T cells whereas IL-17A expression appears later after 5 days (**B-D**). These results suggest that the regulation of IL-26 expression is not as tightly controlled as IL-17A.

4) TGFβ expression by keratinocytes can promote IL-17A production in Th17 cells

By analyzing the expression of TGFβ in the skin of different forms of psoriasis, we observed that TGFβ is more expressed in acute forms of psoriasis (**A**) and is highly expressed by keratinocytes in the skin of erythrodermic psoriasis patients (**B**). Interestingly, TGFβ expression was found to be negatively correlated with IL-26 expression and positively correlated with the expression of IL-17A within acute forms of psoriasis (**C**), suggesting that TGFβ can directly control IL-17A expression in psoriatic skin. To validate this hypothesis, we incubated blood isolated Th17 cells with TGFβ during 7 days. We observed a phenotype shift in Th17 cells with the increase of the IL-17A+ IL-26- population overtime along with a slight decrease of the IL-26+ IL-17A- population (**D-F**). These results suggest that Th17 cells can switch their cytokine production from IL-26 to IL-17A in the presence of TGFβ.

5) IL-26 forms complexes with DNA and induces TGFβ production in keratinocytes

We previously showed that IL-26, a cationic amphipathic molecule, can bind to DNA and form complexes similarly to what other antimicrobial peptides do. These complexes can stimulate pDC and monocytes to produce type I IFNs and IL-6 respectively (**A**). Based on these findings, we tested whether IL-26 can stimulate different skin-resident cells to produce TGFβ. IL-26/DNA complexes, and not IL-26 alone, were found to stimulate keratinocytes to produce TGFβ and to a lesser extent macrophages (**B**).

Summary

References

- Meller S., Di Domizio J., Gilliet M. T(H)17 cells promote microbial killing and innate immune sensing of DNA via interleukin 26. Nat Immunol. 2015 Sep;16(9):970-9