CD200R signalling inhibits *in vitro* Th cell cytokine production

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Background

It is well known that Type 2 T helper (Th2) cells are involved in the pathogenesis of allergic diseases by their release of the signature cytokines, IL-4, IL-13 and IL-5. The inhibitory receptor CD200R was recently identified to be highly expressed on type 2 immune cells, as basophils, ILC2 and Th2 cells. CD200R signalling has been shown to inhibit IgE-mediated mast cell and basophil degranulation, however, the effect of Th2 cells remains unknown. The aim of this investigation was to establish a model system of *in vitro* differentiated Th2 cells and to characterise the influence of CD200R stimulation on *in vitro* differentiated Th cell subsets.

Methods

Naïve human $CD4^+$ T cells were negatively isolated from the PBMCs fraction of healthy individuals. The naïve $CD4^+$ T cells were polyclonal activated with anti-CD3 and anti-CD28. The cells were primed under Th2 (IL-4, anti-IFN-γ and IL-2) conditions for four days and subsequently rested in IL-2 containing medium for three days. At day 7 the $CD4^+$ T cells were split and re-stimulated under Th2 culture conditions with and without the CD200R ligand CD200 (0.1 pg./mL, 10ng/mL and 1000 ng/mL) or a control rhIgG (1000 ng/ml) for up to three days. Cytokine expression was assessed by flow cytometry.

Results

The naïve $CD4^+$ T cells primed under Th2 culture conditions showed a Th2 profile with production of IL-13 (50%) and IL-4 (6%) at day 7 of culture. Interestingly, compared with the control the lowest concentration of the CD200 ligand (0.1 pg./mL), but not the higher concentrations, resulted in a significant decrease in the percentage of IL-13 (50 vs.17%, $p = 0.047$), IL-4 (6 vs.1%, $p = 0.04$) and IL-5 (2 vs. 0.3% $p \leq 0.027$) positive $CD4^+$ T cells at day 1 post re-stimulation. Moreover, relative to the control rhIgG stimulation lower percentages of the Th1 cytokine IFN-γ and Th17 cytokine IL-17A were likewise observed in the 0.1 pg/ml CD200 stimulated $CD4^+$ T cells at day 1 post re-stimulation. Surprisingly, in contrast, to the cytokine production no difference was found in the expression percentage and median fluorescence of the activation marker CD25 when comparing all culture conditions over the stimulation period, indicating that the cells were equally stimulated through the T cell receptor. Finally, no difference in the relative percentage of Th2, Th1 and Th17 cytokine expression were seen between the rhIgG control and CD200 stimulated cultures at day 2 and 3 post re-stimulation.

Conclusion

Altogether, the results indicate that CD200R signalling in *in vitro* differentiated Th2 cells contributes to the inhibition of Th2 but also Th1 and Th17 cytokine production, without affecting the activation of the cells.