INVESTIGATING THE EXPRESSION OF THE HIV-1 CO-RECEPTOR, CCR5, ON HUMAN CD4+ T FOLLICULAR HELPER CELLS.

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Background: CD4+ T follicular helper (Tfh) cells are specialised to provide help to B cells in secondary lymphoid tissue and are critical for regulation of humoral immunity. In humans, germinal center (GC) Tfh cells are defined as CXCR5+PD-1^{high} Bcl6+ CD4+ T cells. Mechanisms behind their infection and accumulation in HIV/AIDS remain unclear. Originally hypothesised to be protected from HIV-1 infection due to the absence of CCR5, Tfh cells have been shown to be productively infected and identified as a major viral reservoir. Recent studies have shown CXCR5- PD-1^{intermediate} precursor Tfh cells express CCR5 prior to differentiation into GC Tfh cells. However, there is currently a knowledge gap about the modulation of CCR5 expression on Tfh cells. We therefore aim to measure the effect of cellular activation and exposure to cytokines and chemokines on CCR5 expression on precursor and Tfh cells to determine if CCR5 expression is transient throughout Tfh cell maturation.

Methods: Multicolour flow cytometry was utilized to establish *ex vivo* expression of CCR5 on human tonsillar Tfh cells (n=3). Pre-Tfh and Tfh subsets were isolated by cell sorting and stained with two different proliferation dyes in order to later track these cells. Labelled pre-Tfh and Tfh subsets were cultured together with unlabeled non-Tfh cells prior to stimulation to induce cellular activation and differentiation; changes in CCR5 and Bcl6 expression were analyzed at three, five and seven days.

Results: Phenotypic analysis demonstrated that a subpopulation of human tonsillar Tfh cells expressed CCR5. An average of 14.93% of Tfh cells were identified as CCR5+ (n=2), suggesting infection with CCR5-using virus is possible.

Conclusion: Preliminary data suggests that the expression of CCR5 on Tfh cells is one of the routes of HIV-1 infection and may be an important factor in reservoir formation. Furthermore, this study will improve understanding of CCR5 expression on human Tfh and pre-Tfh cells and what modulates expression.

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