

## Interaction of type 2 cytotoxic T lymphocytes and mast cell lipid mediators contributes to pathogenesis of eosinophilic Asthma

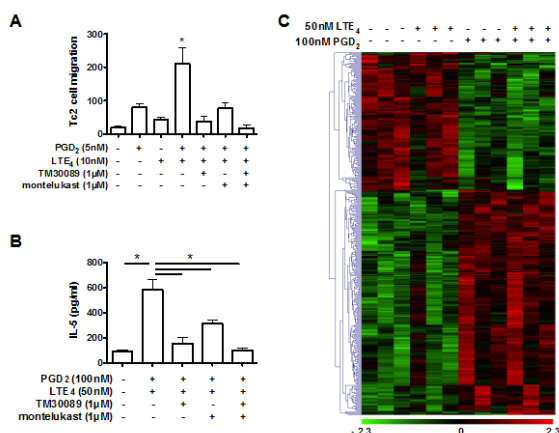
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**Rationale:** Type 2 cytokine (IL-4, IL-5 and IL-13) producing cells in human immune system including type 2 CD4<sup>+</sup> T helpers (Th2), type 2 CD8<sup>+</sup> cytotoxic T cells (Tc2) and group 2 innate lymphoid cells (ILC2) play crucial roles in allergic inflammation including eosinophilic asthma. It has been established that mast cell lipid mediators including prostaglandin D<sub>2</sub> (PGD<sub>2</sub>) and cysteinyl leukotrienes (cysLTs) are strong stimulators to induce type 2 cytokine production in human Th2 and ILC2 cells. Our clinical study has shown significant increase of Tc2 cells and the levels of mast lipid mediators in patients with severe eosinophilic asthma. In this study, we investigated the effects of mast cell lipid mediators in Tc2 cells and how these effects contribute to the pathogenesis of eosinophilic airway inflammation.

**Methods:** Human Tc2 cells were isolated from blood and then cultured for expansion. The effects of PGD<sub>2</sub>, LTE<sub>4</sub> alone or their combination on the cell migration were measured with a chemotaxis assay; and on cytokine production and gene regulation were examined by using microarray, qPCR, ELISA and luminex. The effect of Tc2 activation on the recruitment and survival of eosinophils were assessed by testing eosinophil shape change and apoptosis in response to Tc2 cell conditioned media.

**Results:** PGD<sub>2</sub> and LTE<sub>4</sub> induced migration of Tc2 cells (Fig. 1A), enhanced type 2 cytokine and many other pro-inflammatory cytokine productions in Tc2 cells (Fig. 1B), and broadly regulated the gene transcription in Tc2 cells (Fig. 1C). The combination of PGD<sub>2</sub> and LTE<sub>4</sub> showed additive or synergistic effect on the Tc2 activation. The effects of PGD<sub>2</sub> and LTE<sub>4</sub> in Tc2 cells were abolished by TM30089, a CRTH2 antagonist, and montelukast, a CysLT<sub>1</sub> antagonist, respectively. The supernatant of activated Tc2 cells possesses the capacity to promote eosinophil activation including shape change and survival.



**Figure 1.** *PGD<sub>2</sub> and LTE<sub>4</sub> stimulate Tc2 cell migration (A), IL-5 production (B), and broad gene regulations (C). \*p<0.05*

**Conclusion:** Mast cell lipid mediators PGD<sub>2</sub> and cysLTs play pro-inflammatory roles in Tc2 cell activation. The activation of Tc2 cells could be an important contributor to eosinophilia. Therefore Tc2 cells and mast cell lipid mediator are important targets for therapeutic intervention to control eosinophilic asthma.