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Effects of selective PI3Kδ inhibitors on T cell activation in a murine model

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The Class I phoshoinositide-3-kinase delta (PI3Kδ) isoform is preferentially expressed in leukocytes and regulates key immune functions. Its importance in inflammation signalling, particularly in B and T cells, has stimulated the clinical development of selective PI3Kδ inhibitors. As Th2-driven allergic airway inflammation plays an important role in asthma, our aims was to evaluate the effects of a selective PI3Kδ inhibitor on T cell activation in an ovalbumin (OVA)/ anti-CD3-induced airway inflammation model in transgenic OT.2 mice *in vivo*.

We have developed highly selective PI3Kδ inhibitors that inhibit production of IL-5 in PBMCs and IL-17 in Th17 cells, in a concentration-dependent manner *in vitro*. To elucidate the pharmacodynamic effects of our PI3Kδ inhibitors on effector/memory T helper cells in the lung, we used OVA specific, MHC class II restricted αβ T-cell receptor (TCR) OT.2 transgenic mice(1). Upon local OVA provocation challenge, T-cells were activated and recruited into the lungs. Five days later, the OT.2 mice were given an intranasal (i.n.) anti-CD3 challenge and the downstream effects on PI3K signalling were measured through phosphorylation of S6 ribosomal protein ($pS6RP^{Ser235/236}$), cytokine release and T cell activation markers (CD62L, CD69 and CD25) in absence and presence of a PI3Kδ inhibitor.

We found that $pS6RP^{Ser235/236}$ were significantly downregulated in CD4+T cells by our PI3Kδ inhibitors 6h after anti-CD3 challenge (One-way Anova with Dunnett's multiple comparison test, $p<0.002$). However, no significant effects on activation markers (CD62L, CD69 and CD25) on CD4+ T cells or on cytokine levels in bronchoalveolar lavage fluid were observed at the same time-point.

Our results show that S6RP is phosphorylated at Ser235/236 in CD4+ T cells after anti-CD3 challenge and that this phosphorylation is inhibited by selective PI3Kδ inhibitors. We therefore conclude that $pS6RP^{Ser235/236}$ can be used as a target engagement marker reflecting PI3Kδ activity in this model.

(1) Barnden et al, Immunol Cell Biol. 1998; 76: 34-40