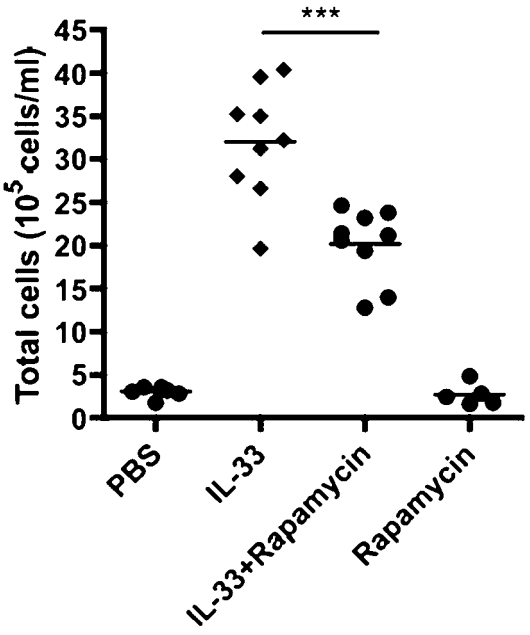
subpopulation of lineage negative innate cells that respond to IL-33 and IL-2.[2] Rapamycin is a macrolide antibiotic that allosterically blocks mTOR, a serine-threonine kinase involved in numerous cellular signalling pathways.

Aim To determine the role of mTOR in IL-33-induced airway inflammation and the effect of rapamycin on IL-33-induced nuocytes in the lung.

Method BALB/c mice were treated with 1 μg of IL-33 intranasally for 5 consecutive days in the presence or absence of rapamycin. Bronchoalveolar lavage (BAL) for cellular and cytokine analysis was performed. Fluorescence-activated cell sorting (FACS) of lung digests were analysed for intracellular IL-5 and cell surface markers.

Results IL-33 induced profound airway cellular infiltration noted in the BAL that was significantly inhibited by rapamycin. Cytokine levels from BAL fluid were also significantly reduced in mice treated with IL-33+ rapamycin. FACS analysis of lung digests demonstrated that IL-33 induced the expansion of lineage negative cells, in keeping with a population of nuocytes, which were the main source of IL-5 in the lung. Furthermore, this population of cells was suppressed by rapamycin.

Discussion Intranasal IL-33 drives mTOR-dependant airway inflammation. Nuocytes are the main source of IL-5 in IL-33-driven airway inflammation. Rapamycin inhibits the production of IL-5 and IL-13 in vivo as well as the expansion of nuocytes in the lung.



Abstract S35 Figure 1 BALB/c mice were treated intranasally with 1 μg IL-33 in the presence or absence of 1 mg/kg rapamycin for 5 consecutive days. The mice were sacrificed on day 6 and BAL total cell counts were performed. ***=p<0.001.

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S36 THE ADHESION RECEPTOR CADM1 PROMOTES HUMAN MAST CELL VIABILITY

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Introduction and Objectives Cell adhesion molecule 1 (CADM1) contributes to cell–cell adhesion, proliferation and adhesion-induced degranulation in human lung mast cells. Previously we found that it is expressed as three alternatively spliced isoforms (major SP4 and SP1, and minor SP6) in human lung mast cells (HLMCs) and only SP4 in the cell line HMC-1, originating from mast cell leukaemia. Here we investigated the role of CADM1 in the viability of HLMCs and HMC-1.

Methods Modulation of CADM1 expression was investigated using adenoviral delivery in both HMC-1 and HLMCs. Cell viability in the absence of survival factors was determined by measuring cell numbers and caspase-3/7 activity.

Results Modulation of CADM1 expression in HMC-1 cells and HLMCs by overexpressing SP4 (exon 8/11) and SP1 (exon 8/9/11) and RNA interference was confirmed by FACS and Western blotting. Overexpression of SP4 did not affect HMC-1 viability (105%±7% of original number of cells) or caspase-3/7 activity (7.7±0.2 FU/cell) in IMDM only for 48 h vs GFP-transduced cells, but overexpression of SP1 reduced cell number (66%±1%) and increased caspase-3/7 activity (9.8±0.3 FU/cell). CADM1 knockdown reduced HMC-1 number (71%±2%) and increased caspase-3/7 activity (9.5±0.2 FU/cell) in these conditions. In contrast to HMC-1 cells, overexpression of SP4 in HLMCs enhanced cell survival (39%±3%) in IMDM alone after 72 h compared to non-transduced cells (31%±1%, whereas downregulation of CADM1 reduced cell number to 26%±2%. Caspase3/7 activity was increased in HLMCs with downregulated CADM1 (99±20 FU/cell) compared to SP4-overexpressing and non-transduced cells (40±4 FU/cell and 20±1 FU/cell respectively). HLMCs displayed lower basal levels of Mcl-1, parallelled with lower survival and higher caspase-3/7 activation compared to HMC-1 cells. CADM1 downregulation in HLMCs coincided with decreased basal expression of Kit and Mcl-1.

Conclusions Modulation of CADM1 isoform expression or CADM1 downregulation in HMC-1 cells reduced survival. Conversely, SP4-overexpression or CADM1 downregulation in HLMCs resulted in increased survival or increased cell death, respectively. CADM1 modulation in HLMCs coincided with modulation of proteins related to survival. Hence, CADM1 promotes survival in human mast cells.

S37 MYD88 DEFICIENCY INFLUENCES MURINE TRACHEAL EPITHELIAL METAPLASIA AND SUBMUCOSAL GLAND ABUNDANCE

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Tracheal epithelial remodelling, excess mucus production, and submucosal gland hyperplasia are features of numerous lung diseases, yet their origins remain poorly understood. Previous studies have suggested that NF-?B signalling may regulate airway epithelial homeostasis. The purpose of this study was to determine whether deletion of the NF-?B signalling pathway protein myeloid differentiation factor 88 (Myd88) influenced tracheal epithelial cell phenotype. We compared wild-type and Myd88-deficient or pharmacologically inhibited adult mouse tracheas and determined that in vivo Myd88 deletion resulted in increased submucosal gland number, secretory cell metaplasia, and excess mucus cell abundance. We also found that Myd88 was required for normal resolution after acute tracheal epithelial injury. Microarray analysis revealed that uninjured Myd88-deficient tracheas contained 103 transcripts that were differentially expressed relative to wild-type and all injured whole tracheal samples. These clustered into several ontologies and networks that are known to functionally influence epithelial cell phenotype. Comparing these transcripts to those expressed in airway progenitor cells revealed only five common genes, suggesting that Myd88 influences tracheal epithelial homeostasis through an