Glucocorticoid receptor α and β expression in bronchial epithelial cells infected with NTHi

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Introduction:

Steroids act through the glucocorticoid receptor (GR), of which the alpha-isoform (GRα) is most abundant. Neutrophilic COPD is associated both with resistance to steroids and with airway bacterial infection, most commonly by non-typeable Haemophilus influenzae (NTHi). We hypothesised that NTHi downregulates GRα or upregulates its inhibitory beta-isoform (GRβ) in bronchial epithelial cells, thereby inhibiting their response to steroids.

Method:

Bronchial epithelial cell line Beas2B were treated with the corticosteroid Fluticasone propionate (0, 1nM or 100nM) for 2 hours prior to infection with $1.5x10^4$, $1x10^6$ and $1.5x10^7$ CFU/ml NTHi (low, medium and high load respectively). 6 hours post-infection supernatants were collected and RNA extracted from cells. RNA was reverse transcribed to cDNA in which levels of GRα, GRβ and GAPDH were determined by SYBR Green PCR and expression calculated using the Pfaffl method relative to untreated cells.

Results:

GRα expression in Beas2B was enhanced by corticosteroid treatment in a stepwise manner for 1nM (median fold increase from untreated: 1.491, IQR: 1.305-1.668), and 100nM (median: 1.742 fold, IQR: 1.51-1.9) ($p>0.05$ for both). Increasing load of NTHi showed no effect on GRα expression. GRβ expression showed little fluctuation from levels of untreated cells upon infection with NTHi alone or high dose corticosteroid, however the two showed a trend to synergistically decrease in GRβ expression when treated with high corticosteroid and high load of NTHi (median fold change from untreated cells: 0.542 (IQR:0.453-0.578) ($p=0.25$) (Figure 1).

Conclusions:

Corticosteroid treatment shows a trend to increased GRα expression on Beas2B cells. Increased NTHi load has no effect on GRα expression of Beas2B cells. GRβ expression appears not to be affected by NTHi infection alone, however with corticosteroid shows a trend to decreased expression. The experiments are to be repeated in primary bronchial epithelial cells to determine whether they follow the trend seen here in a cell line.

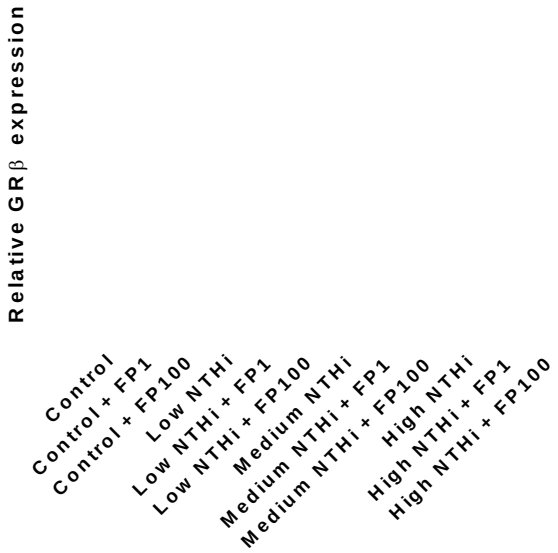

Figure 1: Expression of GRβ in Beas2B cells relative to untreated cells normalised to reference gene GAPDH in a range of conditions.