Distinct Epithelial mRNA and miRNA Expression Profiles During Differentiation and Between EIB(+) and EIB(-) Asthma Phenotypes

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Introduction

✿ Asthma is a respiratory syndrome characterized by periods of reversible airflow obstruction, often in response to inhaled stimuli. In addition to symptoms, there is airway inflammation, structural damage or impaired differentiation. Many of the phenotypic airway epithelial cell features observed in vivo are also evident in vitro, where cells are cultured at an air-liquid interface (ALI) to induce mucociliary differentiation2.

✿ EIB is thought to involve defective epithelial water transport3 and EIB patients have more prominent mediators and increased numbers of basal cells compared to EIB(-) patients4. These findings suggest significant differences between disease phenotypes.

✿ The airway epithelium in asthma has been shown to be remodelled with loss of ciliated epithelial cells, and an expanded basal cell population5, suggesting chronic damage or impaired differentiation. Many of the phenotypic airway epithelial cell features observed in vivo are also evident in vitro, when cells are cultured at an air-liquid interface (ALI) to induce mucociliary differentiation2.

✿ In prior AARBS, we demonstrated that both EIB(+) and EIB(-) AU cultures had impaired mitogenic differentiation, but only EIB(+) had increased numbers of cytokerin-5 expressing basal cells, suggesting epithelial differences in specific asthma phenotypes.

✿ This study aimed to understand the mechanisms of mRNA expression and its effect on mucociliary differentiation of airway epithelial cells from EIB(+) and EIB(-) asthmatic patients.

Methods

Distinct mRNA and miRNA expression profiles are responsible for defective mucociliary differentiation of airway epithelial cells from asthmatic donors with and without exercise-induced bronchoconstriction.

Hypothesis

Specific Aims

1. Perform a global analysis of mRNAs and miRNAs that are differentially expressed between disease groups during differentiation.

2. Identify important mRNA/miRNA interactions that regulate mucociliary differentiation in vitro.

Results

Figure 1. Global analysis of RNA expression during epithelial differentiation in vitro

Figure 2. Summary of differentially-expressed transcripts between donor groups during ALI culture

Figure 3. Biological pathways related to cytoskeleton dynamics and cellular metabolism are aberrantly expressed in asthmatic-derived ALI cultures

Figure 4. MicroRNA-mRNA networks regulate the transition from proliferation to differentiation in ALI culture

Figure 5. (A) The 1344 differentially-expressed genes for group three terms were hierarchically clustered using the complete linkage method into four groups as indicated by numbered boxes as the dendrogram at left. Each row of the heat-map represents a gene, while each column represents a sample and indicates expression (log2) above the median for a given gene. Heat indicates expression below the median. (B) Pathways enriched in each cluster of differentially-expressed genes using GATHER (algorithmic genome data analysis). The rank factor and false discovery rate (FDR) for the enrichment of each pathway are provided. (C) Genetic pathway expression in each cluster are expressed as mean fold change compared to the mean RNA at zero day (plotted horizontal line for cancer (left column), EIB(-) (middle column) and EIB(+) (right column). Genes in each pathway are listed at right.

Acknowledgments

The authors thank the funding bodies for their financial assistance. This project was supported by funding from the Canadian Institutes of Health Research. This grant was approved by the Biostats IGSC.

References