Bacterial lipopolysaccharide (LPS) stimulates IL-8 and granulocyte macrophage-colony stimulating factor (GM-CSF) production in normal human bronchial epithelial (NHBE) cells. Club cell 10-kDa protein (CC10) is an anti-inflammatory protein, produced by epithelial cells, but its role in bronchial airway inflammation is not well established.

Objectives
The aim of this study was to evaluate the effect of CC10 on IL-8, GM-CSF and MUC5AC in NHBE cells stimulated by LPS.

Methods

Cell culture
NHBE cells were differentiated at an air-liquid interface with recombinant human CC10 or vehicle (PBS). Cells were stimulated by LPS and the cell supernatants were harvested on day 14.

IL-8, GM-CSF, MUC5AC secretion
IL-8, GM-CSF, MUC5AC were measured by ELISA. IL-8 and MUC5AC mRNA expression were measured by RT-PCR.

NF-κB and ERK activity
Phosphorylated NF-κB and ERK in whole cell lysates were measured using Western blotting.

Results

Figure 1. CC10 attenuated IL-8 secretion.

Figure 2. GM-CSF and MUC5AC secretion were attenuated by CC10.

Figure 3. CC10 decreased IL-8, and MUC5AC mRNA expression.

Figure 4. CC10 inhibited the phosphorylation of NF-κB and ERK1/2.

Discussion

CC10 may modulate allergic airway inflammation, and decreased concentrations of CC10 are associated with increased severity of inflammatory airway diseases.

We showed that CC10 attenuated IL-8, GM-CSF and MUC5AC production when cells were exposed to LPS. When CC10 was added to the basal culture media, IL-8 secretion was attenuated in both apical and basolateral media. (Figure 1,2)

CC10 blocked LPS-stimulated NF-κB and ERK phosphorylation, and this inhibitory effect of CC10 on cell signaling is consistent with decreased IL-8 and MUC5AC mRNA expression. (Figure 3, 4)

References