**Differential Effects Of β₂-adrenoceptor Agonists On Cytokine Generation From Human Lung Macrophages**

SK Gill, HM Marriott, PT Peachell

*University of Sheffield, Sheffield, UK*

**Introduction:** Lung macrophages are believed to be involved in the pathogenesis of Chronic Obstructive Pulmonary Disease (COPD) and may also have a role in asthma. These inflammatory lung diseases are exacerbated by infections. Currently, treatments for COPD and asthma include steroids such as Dexamethasone as anti-inflammatory agents. β₂-adrenoceptor agonists are also reported to have anti-inflammatory effects but this is disputed.

**Aim:** To investigate the inhibitory effects of long and short-acting β₂-agonists on pro-inflammatory cytokine generation from human lung macrophages in conditions simulating infection (Lipopolysaccharide (LPS)).

**Methods:** Macrophages were isolated from macroscopically normal resected lung tissue of adult participants undergoing surgery mainly for lung carcinoma. Ethical approval and informed written consent were obtained. Cells were purified by Percoll density gradient centrifugation (mean purity 91±2% macrophages, n=6). The cells were incubated overnight, in 24-well culture plates at 2x10⁵ cells/well before challenge with LPS (10 ng/ml) in the presence or absence of short-acting β₂-agonists (Isoprenaline, Terbutaline, Salbutamol) (all 10⁻⁵ M), long-acting β₂-agonists (Formoterol, Salmeterol, Indacaterol) (all 10⁻⁵ M) or the steroid Dexamethasone (10⁻⁷ M). Cell culture supernatants were harvested at 22 h and assayed for TNF-α, IL-6, and IL-8 by ELISA. Statistical significance was determined by 1-way ANOVA followed by Dunnett’s Multiple Comparison test. Significant values were defined as p<0.05.

**Results:** Challenge of macrophages with LPS resulted in high levels of pro-inflammatory cytokine release. TNF-α release was 1,108±271 pg/ml (n=6), IL-6 release was higher at 4,896±1,478 pg/ml (n=5) and IL-8 release was the highest at 55,414±13,729 pg/ml (n=5). Short-acting β₂-agonists showed no significant inhibitory effects on LPS-stimulated cytokine generation. However, the level of TNF-α was significantly decreased by Salmeterol (~49%, p<0.01), Indacaterol (~36%, p<0.05) and the steroid Dexamethasone (~85%, p<0.001, n=6). Inhibition of IL-6 generation followed a similar pattern. The level of IL-6 was decreased by Salmeterol (~56%, p<0.01), Indacaterol (~45%, p<0.05) and Dexamethasone (~79%, p<0.001, n=5). Dexamethasone was the only compound found to significantly inhibit IL-8 generation (~82%, p<0.001, n=5).

**Conclusion:** Macrophages isolated from human lung tissue respond to LPS challenge with the release of high levels of pro-inflammatory cytokines. TNF-α and IL-6 generation is effectively inhibited by some long-acting β₂-agonists (Salmeterol and Indacaterol) but not short-acting β₂-agonists. IL-8 generation is not inhibited by any of the β₂-agonists investigated. Primary macrophages from lung tissue have the potential to be used to investigate novel therapies in lung disease.