Non-Invasive Near-Infrared Fluorescence Quantitative Tomographic Imaging (FMT™) of the Effects of PDE4 Inhibitor Therapy in an LPS Murine Model of COPD In Vivo

Houari Korideck and Jeffrey D. Peterson, PerkinElmer

Abstract

Chronic bronchitis and emphysema, conditions collectively known as chronic obstructive pulmonary disease (COPD), are the fourth leading cause of death in the United States after heart disease, cancer, and stroke. COPD has been associated with the presence of chronic inflammation, generally neutrophilic, that leads to long-term airway obstruction and irreversible, destructive remodeling of lung tissue. An animal model for COPD-like lung inflammation can be established in mice by intratracheal challenge with lipopolysaccharide (LPS), an emulsion of gram-negative bacteria. Current assessments of disease progression rely on invasive measures of pulmonary function and cell infiltration and are therefore limited to terminal assessment. The ability to non-invasively monitor and quantify the underlying inflammatory processes in LPS-induced lung inflammation would provide a significant advance in characterizing longitudinal disease processes and testing potential antilung inflammation agents. We used near-infrared FMT imaging, in combination with fluorescent molecular tomography (FMT) imaging (FMT™), to non-invasively quantify and characterize the disease process and associated edema in the LPS-induced lung inflammation model. In these studies, BALB/c mice received intratracheal instillation of LPS followed by intravenous injection with either the cathepsin-activable fluorescent agent, ProSense 680 (PerkinElmer), to assess alveolar edema and the non-invasively active neutrophils with AngioSense 680 (PerkinElmer) to measure vascular leak in the lungs. This imaging approach provided a robust quantification measurement of two different biological processes. The quantified fluorescence increased correspondingly with dose dependent effects of LPS in bronchoalveolar lavage (BAL) neutrophil counts. The phosphodiesterase IV (PDE IV) inhibitor, Rolipram, when given intraperitoneally at the time of LPS administration, dramatically decreased both neutrophil recruitment and ProSense 680 fluorescence (90%) in the lungs as well as revealing a measurable decrease in the volume of the lung affected by LPS-induced inflammatory edema.

Methods and Materials

Experimental Animals: Specific pathogen-free female BALB/c mice (6-8 weeks old, 18-25 g) were obtained from Charles River (Wilmington, MA) and housed in a specific pathogen-free environment. The animals were provided with water and feed ad libitum.

LPS Inflammation Protocol: LPS-induced inflammation is a simple model for cellular processes occurring in Chronic Obstructive Pulmonary Disease (COPD). On day 0, mice received intratracheal injections of either 0.5 or 10 μg LPS (Escherichia coli K 550, Difco laboratories) or PBS alone. Dose-response and control mice were injected with Pefradex FL (IgG fraction of rabbit serum) or with PBS respectively. Mice were killed on day 3, 6, or 9. The lungs were harvested and sliced into 0.5-mm-thick sections, washed in PBS, and placed in a thermostatically controlled chamber at 37°C. These sections were illuminated with a white light source and the fluorescence signal was imaged at 7 or 24 h for AngioSense and ProSense, respectively.

Results

The LPS-induced lung inflammation is a simple model for cellular processes occurring in COPD. The inflammatory response was characterized by the accumulation of neutrophils in the lungs, as evidenced by the increase in the fluorescent signal produced by the activated ProSense fluorescence. The neutrophil accumulation was dose-dependent, with the highest fluorescence observed in the lungs of mice receiving the highest dose of LPS (10 μg). The fluorescence signal from ProSense 680 was localized to the lung parenchyma and alveoli as compared to the minimal fluorescence detectable in control lungs. This imaging approach provided a robust quantification measurement of two different biological processes. The quantified fluorescence increased correspondingly with dose dependent effects of LPS in bronchoalveolar lavage (BAL) neutrophil counts. The phosphodiesterase IV (PDE IV) inhibitor, Rolipram, when given intraperitoneally at the time of LPS administration, dramatically decreased both neutrophil recruitment and ProSense 680 fluorescence (90%) in the lungs as well as revealing a measurable decrease in the volume of the lung affected by LPS-induced inflammatory edema.

Summary

We demonstrated the ability of the FMT 2500™ in vivo imaging system in combination with two different imaging agents, AngioSense® and ProSense®, to non-invasively visualize and quantify both pulmonary edema and neutrophil activation processes in a murine model of lung inflammation induced by LPS instillation. The consistency of the quantitative tomographic imaging results and its excellent correlation with BAL analysis confirm that FMT imaging is a powerful tool to non-invasively quantify the inflammatory edema and neutrophil activation processes in mouse pulmonary inflammation models.

References


