Hyperpolarized ^{3}He MR imaging of ventilation after bacterial lipopolysaccharide exposure in mice: A model for image-guided sampling of ventilation defects

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Introduction: Hyperpolarized (HP) ^{3}He MRI is a powerful tool for visualizing ventilation defects in human subjects with asthma. The number and size of ventilation defects are known to correlate with the severity of asthma [1]. Though ventilation defects have many postulated causes, their etiology is often unclear. To directly probe the origins of ventilation defects we sought to study a mouse model utilizing lipopolysaccharide (LPS) instillation. LPS inhalation is an important cause of airway inflammation that has been associated with the development of asthma and the severity of various manifestations of the disease [2]. LPS delivered intratracheally to animals is known to cause an acute lung inflammation that results in increased airway resistance and production of T_h1 cytokines within hours of exposure. In this study we created high-resolution ^{3}He MR images to reveal defects associated with LPS-instillation and then used these images to specifically guide tissue sampling to probe for increased concentrations of inflammatory markers, such as cytokines and chemokines.

Methods: C57BL/6 mice (Jackson Labs, Bar Harbor, ME) at 8-10 weeks old were dosed with 50 µl of either LPS (15-50 µg/ animal, N=9) or normal saline (NS) (N=2) via oropharyngeal aspiration following isoflurane induction in accordance with a Duke approved IACUC protocol. A low dead volume 18G tracheostomy tube was inserted after dosing and the animal was placed on a custom HP gas and MR-compatible ventilator. ^{3}He imaging was performed using a 64.8 MHz dual-tuned birdcage coil (L=5.5cm, ϕ=3.5cm) in a 2.0T horizontal 15cm clear bore magnet (Oxford Instruments, Oxford, UK) with shielded gradients (18G/cm), controlled by a GE Excite 12 console (GE Healthcare, Milwaukee, WI). ^{3}He (Spectra Gases, Alpha, NJ) was polarized to approximately 30% in batches of 1.2 liter using a prototype commercial polarizer (IGI.9600.He, MITI, Durham, NC) and dispensed in 200 ml doses for serial imaging. Image time points ranging from 45 minutes to 5 hours post exposure were obtained in order to determine a timecourse of LPS-mediated airway effects. Mice were imaged using a 3D radial acquisition with FOV=2×2×3.2 cm^3 and a matrix of 128×128×32 to give a resolution of 156×156×1000 µm^3. K-space was filled with 11,520 radial views acquired 20 views per breath with TR/TE=5/0.3 ms BW=31.25 kHz either using a fixed flip angle of 13° or a variable flip angle scheme. The 3D images were inspected on a slice by slice basis to identify regions ventilation defects. Defects were visually identified as an area of lung showing decreased intensity relative to surrounding areas, but excluding those portions known to have decreased signal, such as areas immediately surrounding major airways and the most distal edges of each lobe. Defects were then localized to a given lobe of lung and those tissues were dissected out post-mortem and stored at -80C.

Results: With 50 µg LPS, first and second order bronchi constrict resulting in proximal bronchial tree distension, particularly of the right main stem bronchus. Therefore, one marker of LPS effect was the ratio of the width of the right main stem bronchus to the trachea which was 4.1 ± 0.1 at the 50 µg LPS dose, compared to 3.1 ± 0.5 for lower doses of LPS (15, 30 µg) ($p<0.002$) and 3.0 ± 0.5 for NS controls. With lower doses of LPS, less dramatic proximal distension occurs though ventilation defects are often better visualized. Ventilation defects increased in a dose-dependent manner. The mean number of ventilation defects was 3.5 ± 0.7 for 50 µg LPS dosed mice and 1.7 ± 1.1 for lower-dosed LPS mice, while no ventilation defects were visualized for NS dosed mice ($p<0.05$ for 50 µg dose compared to NS, lower dose comparison with NS did not achieve statistical significance). Ventilation defects appear within 2 hours of LPS exposure and remain stable for at least 4 hours post exposure.

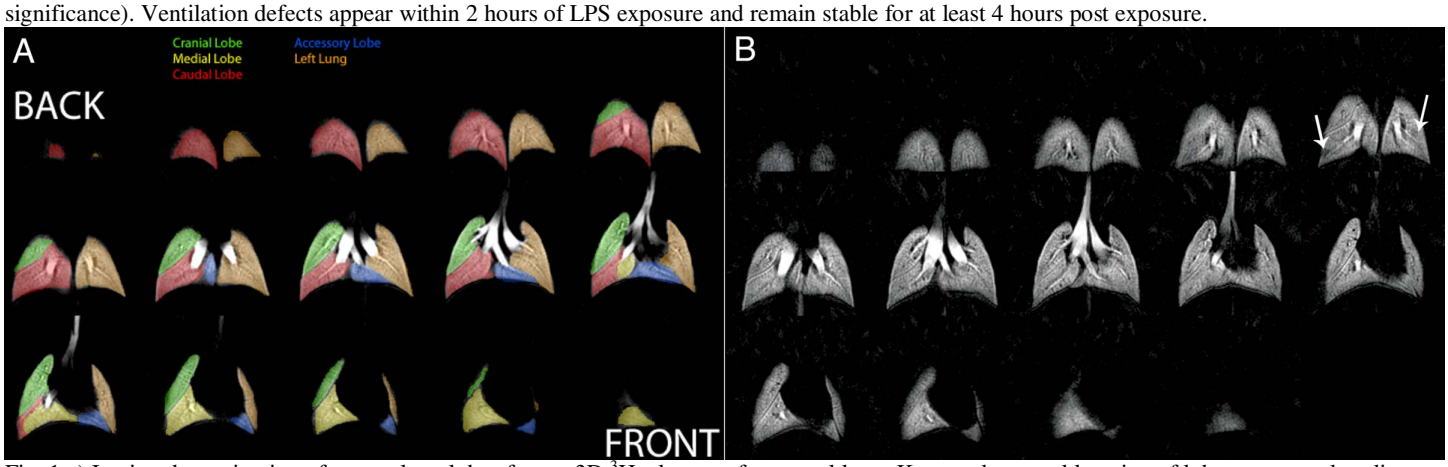

Fig. 1 a) In vivo determination of mouse lung lobes from a 3D ^{3}He dataset of a normal lung. Known shape and location of lobes were used to dissect ventilation defects of LPS mice. b) 3-hr timepoint after 30 µg LPS dose. Arrows point to ventilation defects.

Conclusions:

HP ^{3}He MRI imaging of LPS dosed mice appears to be a suitable model for visualizing ventilation defects. This is the first study to use HP ^{3}He MRI imaging to visualize regional ventilation differences resulting from LPS inhalation in mice and to sample the ventilation defects based on imaging. The presence of ventilation defects is dose-dependent and defects occur within 2 hours of LPS dosing. Planned analysis of tissue samples dissected with guidance from HP ^{3}He MRI should be useful in determining whether ventilation defects contain local increases in inflammatory markers.

References:

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