# Diffusion Imaging with Hyperpolarized Helium MR in Surfactant Protien C Deficient Mice

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## INTRODUCTION:

Diffusion MRI of hyperpolarized noble gases provides a noninvasive and sensitive means of measuring changes in regional lung microstructure (1). Values for the gas apparent diffusion coefficient (ADC) are expected to be sensitive to changes in lung microstructure, particularly alveolar size. This methodology can be applied to humans as well as animal models of lung disease, though its application to small animals, particularly mice, poses technical challenges. These challenges include the need to endotracheally intubate small animals, the necessity of delivering hyperpolarized gas through a specialized ventilator in the presence of a strong magnetic field, and the small alveolar size of mice relative to the mean square diffusive displacements of $^3$He gas. As proof of principle, we have employed this method to evaluate a transgenic mouse model of surfactant protein C (SPC) deficiency. SPC enhances the stability and spreading of phospholipids by recruiting phospholipids to monolayers and multilayers at the air-liquid interface of the alveoli. The SPC-/- mutation is associated with idiopathic interstitial pneumonitis in humans, an autosomal dominant trait. SPC-deficient mice have a progressive, nonhomogenous lung disease with inflammation, loss of alveolar structure, and fibrosis (2). The SPC-deficient animals employed for this study were evaluated at age 8-10 months, a time at which severe pulmonary fibrosis would be expected to reduce alveolar size, leading to a decrease in the ADC of $^3$He. If changes in alveolar structure can be detected by diffusion maps, this could provide a tool to monitor progression of bronchopulmonary displasia in prematurely-born infants. Such a tool might aid making treatment decisions, assist with predicting long term outcome, and provide further insight into understanding this neonatal pulmonary disease processes.

## MATERIALS AND METHODS:

We obtained SPC-deficient knockout mice generated from a congenic 129/Sv strain from the division of Pulmonary Biology at Cincinnati Children's Hospital Medical Center. The mice were anesthetized with xylazine/ketamine mixture. They were intubated with a 1.27-mm endotracheal tube passed over a guide wire through the vocal cords. An RF coil, tunable to both the hydrogen and $^3$He resonance frequencies, was placed around the mouse's chest. The endotracheal tube was connected to an MRI- and $^3$He-compatible small animal ventilator made by the Washington University physics department (4). The ventilator was set at a rate of 120 breaths per minute, and 10 ml/kg of gas ($^3$He and $O_2$) was delivered. Inhalation time was set at 4.7. Data were obtained using a Varian 4.7-T imaging system. A 2D gradient echo multislice pulse sequence was used to acquire 1- or 2-mm thick image slices with a field of view (FOV) 4 cm x 4 cm and spatial resolution of 0.31 mm x 0.31 mm. Maps of the ADC were made by acquiring two interleaved images, one with $b = 0$ and one with $b = 5.78$ s/cm$^2$. The diffusion time was 533 µs. ADC values were calculated on a pixel by pixel basis using monoexponential signal decay function, excluding signal from the trachea.

## RESULTS:

Two control and four SPC-deficient mice were imaged at 8 to 10 months of age. The typical signal to noise ratio was 30. Figure 1 shows coronal ADC maps from SPC-deficient (A) and control (B) mice. There is little regional variation in ADC values for either animal. Standard deviations for ADC values within the images were on the order of 0.06 cm$^2$/s. The ADC of the SPC-deficient mice is 0.127 ± 0.020 cm$^2$/s ($n$ = 4). The ADC values of the two control mice were 0.144 and 0.181 cm$^2$/s. Histograms of ADC values from single animals are shown in Figure 2.

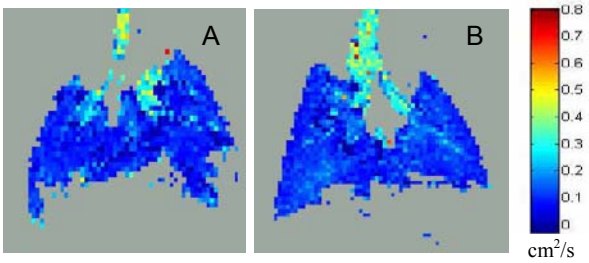


Figure 1: $^3$He ADC map of SPC-deficient (A) and control (B) mouse lung. The lung in figure A is smaller due to fibrosis.

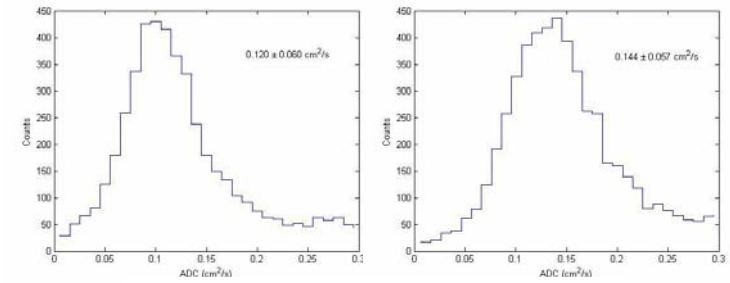


Figure 2: Histogram showing the ADC for a SPC-deficient mouse on the left and a control mouse on the right.

## CONCLUSION:

Measurement of $^3$He ADC values is feasible in mouse lung. It is worth noting that the alveolar size of a 33-g mouse is 58 ± 4 µm, as compared with an alveolar size of 225 ± 15 µm in humans (4). Given the relatively high free ADC value for $^3$He gas (on the order of 0.88 cm$^2$/s), we employed the shortest diffusion time possible with our hardware (533 µs) in an effort to maintain mean square displacement values for $^3$He on the order of the alveolar size of mice. With this diffusion time, the mean square gas displacements for this study were ~120 µm, suggesting that a given $^3$He molecule sampled more than one alveolus during the diffusion measurement. Using these acquisition parameters, we were able to detect differences between control and SPC-deficient mice, with a trend towards lower ADC values in affected mice, consistent with the effects of pulmonary fibrosis. This indicates that hyperpolarized helium diffusion measurements are sensitive to the chronic pathological changes associated with SPC deficiency.

## REFERENCES:

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