Quantitative Assessment of Lung Microstructure in Healthy Mice using Lung Morphometry with Hyperpolarized 3He Diffusion MRI

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Introduction: The lung morphometry technique with hyperpolarized ^{3}He MRI [1, 2] allows in vivo quantification of lung geometrical parameters at the acinar level. In humans imaging provides results similar to direct morphological measurements. In this technique, lung acinar airways are treated as cylindrical air passages with radius *R* covered inside with alveolar sleeve of depth *h* [3]. Knowing these parameters allows estimation of standard histological parameters such as mean chord length *Lm*, surface-to-volume ratio *S/V*, and density of alveoli per unit lung volume N_a [2]. The premise of the introduced lung morphometry technique is establishing the relationship between diffusion attenuated MR signal and lung geometrical parameters *R* and *h*. Herein the lung morphometry technique was applied to study lung microstructure in mice.

Methods:

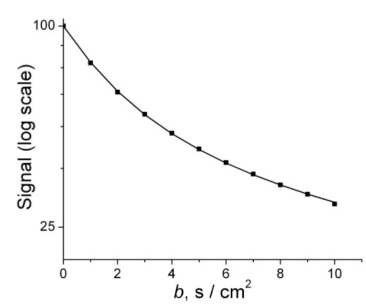
Gas Preparation: A commercial 9600.He polarizer (General Electric, Fairfield, CT) was used to produce ~1 L of ^{3}He gas at ~40% polarization level. Prior to experiments the gas was transferred to a 1 L Tedlar bag, which was kept in the fringe field during the course of several experiments on one mouse lung.

Lung Preparation: Six mice were used for this study. All experiments were approved by the Washington University Animal Study Committee. We used lungs excised from freshly sacrificed mice (C57BL/6N, males, 3-4 months old, 21-27 grams). Mice were euthanized using 0.1 ml of Ketamine and lungs were extracted. The trachea was tied around a plastic needle to ensure a tight seal. Using a setup consisting of six valves, lungs were repeatedly purged with ^{3}He to dilute oxygen and other gases out of the lungs. HP ^{3}He was brought to the fringe field of the magnet. Using the same 6-valve setup, any residual oxygen was purged. ~850 µl HP ^{3}He gas delivered to the lungs, and topped with ~150 µl of N_2 to eliminate ^{3}He MR signal contribution from the non-acinar portion of the lung. The Tedlar bag is disconnected and the lungs are scanned. Besides keeping the oxygen out, the main objectives of the 6-valve system are repeatable delivery of HP ^{3}He at the consistent lung inflation and a precise topping with N_2 blanket.

MR Experiment: The data were collected on a Varian 4.7T scanner with 120 mm horizontal bore equipped with 60 G/cm gradients. The typical diffusion time used in human experiments [1, 2] is 1.8 ms, which was small enough for the ^{3}He molecule to remain inside of a single acinar airway. In mice, the airways are much smaller, hence we have to use much shorter diffusion time. For that reason, the bipolar diffusion-sensitizing gradients ($\Delta = \delta = 0.44$ ms), with fixed ramp times ($\tau = 0.175$ ms) and eleven b-values ranging from 0-10 s/cm^2 were used. Gradients were applied in three orthogonal directions and signals were averaged together. Data were analyzed using relationship between MR signal and lung microstructure that was established for mouse lung (results to be published elsewhere) using Monte Carlo simulations similar to those presented in [4] for human lungs.

Results: A typical dependence of the log MR signal on the b-value is shown in Figure on the right (symbols) along with the fitting curve (solid line). Results demonstrate an excellent fit with low χ^2, about 0.05. A very important feature of the signal dependence on the b-value seen in Figure is a substantial deviation from mono-exponential behavior, as predicted by our model. The excellent fitting of our model to the signal substantially adds to model validation. The obtained geometrical parameters, *R* and *h* along with *Lm* calculated per [2], for all six mice are shown in the Table at left. Our result for mean chord length, *Lm* is consistent with literature data. Indeed, *Lm*=44.7±2.9µm was reported in [5] for 6-8 weeks old mice, and *Lm* = 52±1µm was found in [6] for 10-12 weeks old mice, while we obtained *Lm* = 62±12 µm for 13-17 weeks old mice.

mouse	*R, µm*	*h, µm*	S_a/V_a, cm^{-1}	N_a, mm^{-3}	*Lm, µm*
1	103.7	63.1	738.34	2983.6	54.2
2	107.8	48.3	554.48	2655.9	72.1
3	84.1	46.3	836.44	5593.5	47.8
4	119.1	64.1	579.71	1969.4	69.0
5	109.3	46.2	523.72	2548.1	76.4
6	94.3	58.0	787.55	3634.2	50.8
mean	103.05	54.33	670.04	3230.8	61.72
std	12.29	8.39	133.48	1280.2	12.21



Conclusion: Results obtained in this study demonstrate that the ^{3}He lung morphometry technique can be used to quantify lung morphometrical parameters in mice.

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