Dynamic Hyperpolarized 129 Xe MR Spectroscopy of rat lungs inflamed by Stachybotrys chartarum spores.

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Introduction

Several studies have reported the use of hyperpolarized ^{129}Xe (HXe) MR spectroscopy as a probe of lung function. In addition to the alveolar gas phase resonance, Sakai et al found three dissolved phase resonances in rat lung, which were attributed to lung parenchyma, red blood cells, and plasma/adipose tissue (1). Ruppert et al exploited the large chemical shift difference between the gas phase and the dissolved phase resonances to investigate the time course of xenon gas exchange between the lung tissue and the alveolar gas. By selectively saturating the xenon signal from the lung tissue resonance, allowing a variable time delay, and then obtaining spectra with another pulse selective for the lung tissue, it was possible to characterize the exponential recovery of magnetization in the lung tissue by a transfer time (2).Using this dynamic HXe spectroscopy technique, Mansson et al have demonstrated prolonged transfer times in rats with lung inflammation caused by bacterial lipopolysaccharide compared to control animals (3). Dynamic HXe spectroscopy thus seems promising for the detection of subtle gas exchange abnormalities associated with inflammation of the lung.*Stachybotrys chartarum* is a widespread environmental mold, spores of which contain a number of toxins and have been shown to provoke an inflammatory response in rat lungs (4). Previous results have shown narrowing of the adipose peak and a shift towards the lung peak in rats with severe spore induced lung inflammation(5).That work is extended in the present study to determine whether dynamic HXe spectroscopy would demonstrate changes in gas transfer times in rats instilled intratracheally with *S.chartarum* spores, and to what degree these changes correlate with histological grade of inflammation.

Materials and Methods

Adult male Wistar rats (300-350g)were instilled intratracheally with 350 microlitres of fungal spores ($1.4X10^6$/ml) in saline. After approximately 24-36 hours the rats underwent tracheostomy and mechanical ventilation under isoflurane anesthesia. Hyperpolarized xenon (natural abundance, 26% 129Xe) was produced by optical pumping with rubidium vapor in a custom made polarizer cell and accumulated for 5 minutes at a flow rate of 3 ml /min as xenon ice at liquid nitrogen temperature. After sublimation in a water bath, the xenon was administered as a single bolus inspiration to the rat , using a pressure release valve set to 100 mbar above atmospheric pressure to avoid barotrauma.Spectra were acquired as previously described(5). Six controls and six spore instilled rats were studied. Five spectra from each rat using five separate gas boluses were signal averaged and the peak areas corresponding to lung tissue were normalized to the gas peak of the preceding spectrum (to remove T_1 and RF saturation effects on the gas peak) and plotted against delay time τ. These data were fitted to the model of Mansson et al(3) to obtain the transfer time constant T_{tr}. The pathological changes in the hematoxylin/eosin fixed lungs were graded on a scale of 1-5 based on reduction of alveolar space by inflammatory changes at sites of spore impaction (grade 1: less than 10% reduction, grade2:10-25% , grade 3: 25-50%, grade 4: 50-75%, grade 5:greater than 75%).

Results

Representative transfer curves are shown in Figure 1.The model fitted is $y=C_1(1-\exp(-x/T_{tr}))+C_2x$ (3).Table 1 shows the transfer times and inflammation grade for each rat . The control group had a shorter mean T_{tr} than the spore instilled group (20 +/- 3 ms vs 35 +/-8 ms. While the control group fell in the range 16 ms to 24 ms, there was some variability in the experimental group, with transfer times ranging from 43 ms to 24 ms.

Figure1. Representative transfer curves for spore and control rat

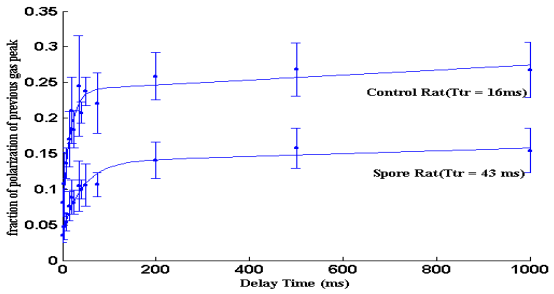

Table 1. Transfer times for lung parenchyma (196 ppm) for control and fungal spore instilled rats. Histological grade refers to diseased animals.

Control Rat	Transfer Time	Spore Rat	Transfer Time	Histological Grade
1	16 ms	1	43 ms	2
2	24 ms	2	40 ms	3
3	17 ms	3	30 ms	4
4	20 ms	4	28 ms	1
5	23 ms	5	24 ms	4
6	22 ms	6	42 ms	4
Mean+/-S.D.	20+/-3 ms	Mean+/-S.D.	35+/-8 ms	

Discussion and Conclusion

These experiments permitted detection of gas exchange abnormalities in the lungs of rats exposed to *S. chartarum* spores, as evidenced by increased transfer times for the lung tissue compartment. The degree of change was similar to that reported by Mansson et al(3) in the LPS model of rat lung inflammation, where the T_{tr} values were 29 +/- 4 ms and 40+/-5 ms for control animals and experimental animals.respectively. The difference between the two control groups suggests that the technique of gas administration may influence the T_{tr} . Mansson et al administered a fixed tidal volume of 1ml/kg whereas the current experiment used a constant pressure; the volume of gas entering the lung was likely different. Diffusing capacity for CO(DL_{CO}) is known to be higher at lung volumes closer to total lung capacity(6). As a measure of gas diffusion, T_{tr} may have a similar dependence.Precise establishment of the lung volume at which a particular T_{tr} is measured may be of importance in comparing T_{tr} values from different experiments. There was no obvious correlation between the degree of *S.chartarum* spore induced histological inflammation and T_{tr} ,possibly due to the inhomogeneous distribution of the granulomatous changes in this disease model . This work shows that dynamic HXe spectroscopy is capable of detecting changes in T_{tr} caused by *S. chartarum* spore inhalation; the dependence of T_{tr} on lung volume and pressure is a subject for further study.

References

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