# Single lobe emphysema induction in the rat lung detected with diffusion-weighted 3He-MRI

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**Introduction:** Diffusion weighted imaging yielding the apparent diffusion coefficient (ADC) is among the main applications of lung imaging with hyperpolarized noble gases. Because ADC values are related to molecular mobility, they are expected to reflect changes in lung microstructure, and hence should be of use as a detector for lung diseases. It has been found that ADC can distinguish between healthy and diseased subjects in elastase induced models of emphysema in rats [1, 2] and in naturally developed emphysema in humans [3, 4]. However, a thorough understanding of all factors determining ADC values remains an elusive goal.The main aim of this work was to determine the best parameters to differentiate between control and emphysematous lungs by ADC measurements and to demonstrate feasibility of instilling elastase in only one lung. A special ventilation protocol was used to detect differences between emphysematous and healthy lung tissue. The protocol consisted of ventilating and imaging the animals with two different gas mixtures with similar diffusion coefficient. The utilized gas mixtures make ADC measurements independent of the gases concentration of the final mixture in the lungs.

**Methods:** In ten Wistar rats the left lung was instilled with 25 units of porcine pancreatic elastase dissolved in saline. The applied volume was 0.1ml/100g animal weight. Right lung was left as an internal control. The whole cohort was measured four weeks (4W) after the instillation and four of the animals were additionally imaged two weeks after the first measurement (6W). Prior to imaging, animals were anaesthetized with thiopental sodium, intubated and connected to a mechanical respirator-applicator, which ventilated the animals at 60 bpm with a mixture consisting of 21% $O_2$ and 79% $^4$He. The same ventilator was used to apply a bolus of HP-gas. In the acquisition mode a 10s-breath-hold was applied to the animals. Animals were imaged during the full-inspiration breath-hold at ~23mbar (BH) and also at the end of the expiration period (EEV). EEV was preceded by a short full-inspiration. The gas applied for imaging consisted of 77% HP$^3$He and 23% $N_2$. This mixture and the one used for ventilating the animals prior to MRI had a very similar free intra-diffusion coefficient. We used these gas mixtures to reduce the effect on the ADC results due to the different diffusivity of mixtures with either different gases composition or different gases concentration.
The image data was acquired on a Bruker Biospec 70/20 spectrometer operated at 0.5T. A radial sequence was used to obtain diffusion images with 4 b-values (0.05, 0.707, 2.11 and 4.26 s·cm$^{-2}$) and diffusion time of 1.5ms.

**Results:** Figure 1 shows the ADC mean values obtained from the left (elastase-treated) lung and the right (control) lung of the rats measured under both ventilation conditions, BH and EEV at 4W. The error bars represent the standard deviation of ADC of each animal group. To avoid contributions of larger airways the calculation of the ADC values was only carried out over the lung peripheral areas. A significant difference between the ADC values of the two lungs at the end of the expiration ($P < 0.001$) with an increase for the emphysematous lung was observed. This effect was not seen when animals were imaged at full-inspiration BH. A Student´s t-test did show no significant differences in ADC between measurements at 4 and 6 weeks in any of the lungs in neither ventilation conditions. Figure 2 shows a representative ADC map obtained at the end of the expiration. A PEEP versus expiration curve for both lungs is shown in Figure 3. The emphysematous lung shows always higher values than the healthy one, being the differences more pronounced at low pressures. Above certain expiration pressure (~6mbar), the ADC reaches a plateau in both lungs.

**Discussion:** Using the healthy lung (left on the ADC map) as an internal control removes the effect of inter-subject variability and also enables the visualization of differences between the elastase-treated and non-treated lung. The procedure of instilling elastase in only one lung proves to be reliable and efficient. This allows the reduction of the total number of animals used in the experiment while preserving the statistics and increasing sensitivity. High residual volume and alveolar expansion are typical for the emphysematous damage produced by the elastase. Measuring at the end of the expiration the greater ADC value observed in the elastase-treated lung compared to the control one may directly be related to the higher residual volume in the emphysematous lung. On the other hand, at full-inspiration conditions the similar ADC values obtained in both lungs could be explained as a consequence of the relative high pressure applied which may equalize both lungs. No significant differences between the measurements at 4 and 6 weeks for any of the lungs were observed, indicating that the damage caused by the elastase was fully developed after four weeks. We conclude that the characteristics of emphysematous lungs (higher airspaces, higher compliance and higher residual volume) are visible and easier to detect at EEV conditions, once the contribution of other factors, such as different gases with different diffusion coefficients have been minimized by using the proposed gas mixture.

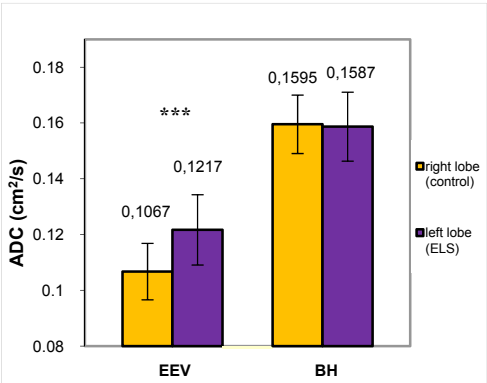


**Figure 1:** ADC mean values of the control and elastase-treated lungs of ten animals imaged at full-inspiration breath-hold of 25mbar (BH) and at the end of the expiration (EEV) four weeks after elastase instillation.

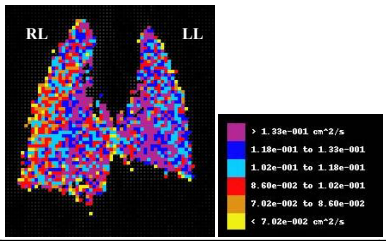


**Figure 2:** ADC map of one animal obtained in EEV. Highest values can be seen in purple, whereas the lowest ones are shown in yellow.

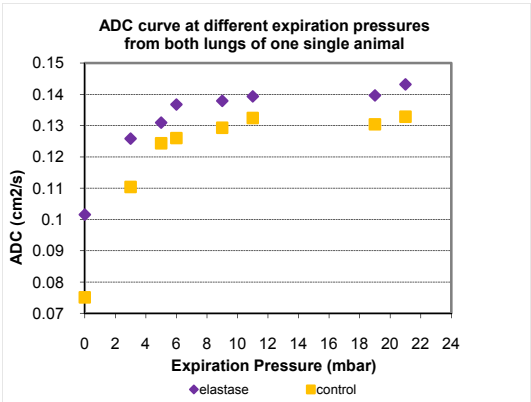


**Figure 3:** ADC versus expiration pressure of one representative animal. ADC values for the emphysematous lung were higher than for the control one at each pressure. At low pressure the differences in ADC between both lungs were higher.

**References: [1]** Chen XJ *et al*. PNAS, 2000; 97(21): 11478-11481, **[2]** Peces-Barba G *et al*. E. Respir. J., 2003; 22: 14-19, **[3]** Saam BT *et al*. Magnetic Resonance in Medicine, 2000; 44: 174-179, **[4]** Salerno M *et al*. Radiology, 2002; 222: 252-260. **Acknowledgments:** Supported by the Marie-Curie training network MRTN-CT-2006-03602, PHeLINet.