Noninvasive Monitoring of the Response of Human Lungs to Low-Dose Lipopolysaccharide Inhalation Challenge Using MRI: A Feasibility Study

Agilo L. Kern, MSc,1,2 Heike Biller, MD,2,3 Filip Klimeš, MSc,1,2 Andreas Voskrebenzev, PhD,1,2 Marcel Gutberlet, PhD,1,2 Julius Renne, MD,1,2 Meike Müller, PhD,2,3 Olaf Holz, PhD,2,3 Frank Wacker, MD,1,2 Jens M. Hohlfeld, MD,2,3,4 and Jens Vogel-Claussen, MD1,2*

Background: Development of antiinflammatory drugs for lung diseases demands novel methods for noninvasive assessment of inflammatory processes in the lung.

Purpose: To investigate the feasibility of hyperpolarized $^{129}$Xe MRI, $^1$H T1 time mapping, and dynamic contrast-enhanced (DCE) perfusion MRI for monitoring the response of human lungs to low-dose inhaled lipopolysaccharide (LPS) challenge compared to inflammatory cell counts from induced-sputum analysis.

Study Type: Prospective feasibility study.

Population: Ten healthy volunteers underwent MRI before and 6 hours after inhaled LPS challenge with subsequent induced-sputum collection.

Field Strength/Sequences: 1.5T/hyperpolarized $^{129}$Xe MRI: Interleaved multiecho imaging of dissolved and gas phase, ventilation imaging, dissolved-phase spectroscopy, and chemical shift saturation recovery spectroscopy. $^1$H MRI: Inversion recovery fast low-angle shot imaging for T1 mapping, time-resolved angiography with stochastic trajectories for DCE MRI.

Assessment: Dissolved-phase ratios of $^{129}$Xe in red blood cells (RBC), tissue/plasma (TP) and gas phase (GP), ventilation defect percentage, septal wall thickness, surface-to-volume ratio, capillary transit time, lineshape parameters in dissolved-phase spectroscopy, $^1$H T1 time, blood volume, flow, and mean transit time were determined and compared to cell counts.

Statistical Tests: Wilcoxon signed-rank test, Pearson correlation.

Results: The percentage of neutrophils in sputum was markedly increased after LPS inhalation compared to baseline, $P = 0.002$. The group median RBC-TP ratio was significantly reduced from 0.40 to 0.31, $P = 0.004$, and $^1$H T1 was significantly elevated from 1157.6 msec to 1187.8 msec after LPS challenge, $P = 0.027$. DCE MRI exhibited no significant changes in blood volume, $P = 0.64$, flow, $P = 0.17$, and mean transit time, $P = 0.11$.

Data Conclusion: Hyperpolarized $^{129}$Xe dissolved-phase MRI and $^1$H T1 mapping may provide biomarkers for noninvasive assessment of the response of human lungs to LPS inhalation. By its specificity to the alveolar region, hyperpolarized $^{129}$Xe MRI together with $^1$H T1 mapping adds value to sputum analysis.

Level of Evidence: 1
Technical Efficacy Stage: 2
AIRWAY INFLAMMATION is believed to play a vital role in the pathogenesis of many lung diseases. Specifically, progression of chronic obstructive pulmonary disease is associated with neutrophilic airway inflammation. Segmental challenge of the lung using lipopolysaccharide (LPS) causes a local neutrophilic inflammatory response, thereby providing a disease model for development of targeted antiinflammatory drugs. Cell counts from bronchoalveolar lavage then enable a quantification of the inflammatory response for treatment monitoring. However, these procedures entail repeated bronchoscopies and thus cause additional burden to the subjects.

An alternative approach to segmental challenge is low-dose LPS inhalation, causing a mild inflammatory response in the whole lung. Collection of induced sputum is generally well-tolerated and inflammatory cell counts then similarly enable quantitative assessment of inflammation for testing the efficacy of investigational new drugs. However, the origin of sputum from the airways is less controlled and the potential of sputum analysis for accurate disease monitoring at the alveolar level is thus limited. Further, repeated sputum induction has been shown to alter cell counts at later times, which limits its use for repeated assessments.

Magnetic resonance imaging (MRI) provides spatially resolved diagnostic information and can be repeated without detrimental effects. It has been shown that 1H turbo inversion recovery magnitude (TIRM) MRI and T1 mapping MRI are sensitive to airway inflammation after segmental allergen challenge in asthmatics. However, TIRM MRI only provides semiquantitative information on local fluid content, and 1H T1 is not very specific for a certain pathology.

MRI and spectroscopy of inhaled hyperpolarized 129Xe enable noninvasive assessment of gas uptake to lung parenchyma and blood and yield specific, quantitative information on lung function at the alveolar level. Models of 129Xe uptake dynamics have been developed, providing physiologic parameters like septal wall thickness, surface-to-volume ratio, and capillary transit time of blood. The specificity of parameters derived from 129Xe dissolved-phase MRI and spectroscopy to lung structure and function at the alveolar level suggests that changes in these parameters could be quantitatively associated with the degree of inflammation in the alveolar region and could potentially be used as surrogate markers, for example, for cell counts.

The purpose of this study was to test the feasibility of hyperpolarized 129Xe, 1H T1 mapping and dynamic contrast-enhanced (DCE) MRI for monitoring the response of human lungs to inhaled LPS challenge. The potential of MRI-derived biomarkers for noninvasive assessment of inflammation was tested by comparison to inflammatory cell counts from sputum analysis.

Materials and Methods

Study Design

This prospective study (ClinicalTrials.gov Identifier NCT03044327) was approved by the Institutional Review Board and all subjects gave written informed consent. The study was conducted at a single research center between February and November 2017 and included inhaled LPS challenge of the lung. MRI was performed before and 6 hours after inhaled LPS challenge. The timepoint for MRI after LPS challenge was chosen to coincide with the time of maximal inflammatory response in terms of cell counts, although there is still some controversy about this point.

Subjects were included if they had normal lung function (forced expiratory volume in 1 sec [FEV1] >80% of predicted normal, FEV1/forced vital capacity [FVC] >70%) and were nonsmokers with a smoking history of less than one pack-year. Exclusion criteria were clinically relevant history of allergies, elevated level of immunoglobulin E, lower respiratory tract infections within 4 weeks of the first MRI, and general MRI contraindications.

LPS CHALLENGE AND CELL COUNTS. For inhaled LPS challenge, 2 μg LPS was dissolved in sterile water and administered using a breath-controlled nebulizer. Inspiration was adjusted to a low flow rate of 150 mL/s, which is expected to improve deposition of LPS to the alveoli. Sputum was induced by inhalation of nebulized saline and immediately processed.

Magnetic resonance imaging (MRI) was performed at 1.5T (Magnetom Avanto; Siemens Healthcare, Erlangen, Germany). 129Xe MRI was performed using a custom-made birdcage transmit and 16-channel receive coil (Rapid Biomedical, Rimpar, Germany).

Transmitter calibration was performed using a dedicated pulse sequence in a separate breath-hold. Subjects inhaled 1 L of a gas mixture containing 0.3 L hyperpolarized xenon with natural isotope ratio and 0.7 L nitrogen for this purpose.

A 3D-radial multiecho sequence and a 3-point Dixon algorithm were used for interleaved imaging of the hyperpolarized 129Xe dissolved phase, ie, 129Xe in red blood cells (RBC) and tissue/plasma (TP), and gas phase (GP). Sequence parameters were repetition time 19 msec, echo times 0.74/2.36/3.98 msec for dissolved phase, 0.74/2.36 msec for gas phase, flip angle 23° for dissolved phase / 0.4° for gas phase, reconstructed resolution 7.6 × 7.6 × 17.6 mm, whole-lung coverage, scan time ~10 sec. Spectroscopy of the dissolved phase (~18 Hz resolution), ventilation imaging (stack-of-stars gradient-echo sequence, 2.4 × 2.4 × 17.6 mm resolution, 14 coronal slices, scan time ~6 sec) and a fast 1H scan (gradient echo sequence, reconstructed resolution 2.0 × 2.0 × 5.0 mm, scan time ~3 sec) were also performed during this second breath-hold. For these acquisitions, subjects inhaled a 129Xe gas mixture of volume 1/3 FVC, starting from residual volume, containing 1 L hyperpolarized xenon with isotopically enriched 129Xe fraction (Linde, München,
Germany) and air similarly as in previous studies.\textsuperscript{15} \textsuperscript{129}Xe and air were kept in two different Tedlar bags of 1 L capacity (Jensen Inert Products, Coral Springs, FL) for this purpose and mixed only during inhalation by the subject.

Dynamic gas-uptake measurements of \textsuperscript{129}Xe were made using a chemical shift saturation recovery (CSSR) spectroscopy sequence.\textsuperscript{16} Spectra were acquired in a third breath-hold after inhalation of a gas mixture containing 0.5 L hyperpolarized \textsuperscript{129}Xe and 0.5 L nitrogen from a 1 L Tedlar bag and subsequently room air to achieve full lung inflation. The spectroscopic bandwidth was $\pm 16.7$ kHz at 32.6 Hz resolution; sampled delay times ranged from 3–700 msec, resulting in a measurement time of ~9 sec. Saturation of the dissolved phase was performed using a 2.4 msec long rectangular 90° pulse at 198 ppm. The residual \textsuperscript{129}Xe magnetization was used for ventilation imaging in the same breath-hold as before.

\textsuperscript{1}H T\textsubscript{1} maps were obtained in typically 8–9 slices covering the lung during breath-hold at end-tidal inspiration with the central slice at the tracheal bifurcation. An inversion recovery fast low angle shot sequence\textsuperscript{2} was used for imaging, repetition/echo time 3/0.66 msec, flip angle 8°, resolution 3.9 × 3.9 mm, slice thickness 15 mm, no gaps, 32 inversion times (103–6055 msec).

DCE MRI was performed after intravenous injection of 0.033 mmol/kg bodyweight gadopentetate using a time-resolved angiography with stochastic trajectories sequence\textsuperscript{17} with repetition/echo time 2.37/0.8 msec, voxel size 2.0 × 2.0 × 5.0 mm, temporal resolution ~1.3 sec, 30 frames.

### Data Analysis

The MRI reader was blinded to the results of induced sputum analysis. For computation of whole-lung dissolved-phase ratios in \textsuperscript{129}Xe dissolved-phase MRI, an automatically determined mask based on signal-to-noise ratio (SNR) thresholding was applied to the data. This mask excluded voxels with SNR less than 5 in the first echo of either gas or dissolved phase. Quantification of ventilation defect percentage was performed using a mask of the thoracic cavity obtained by application of a region-growing algorithm in the \textsuperscript{1}H gradient echo images and signal binning as described previously.\textsuperscript{18}

The masks for \textsuperscript{1}H T\textsubscript{1} mapping and DCE MRI were similarly determined by application of a region-growing algorithm within the thoracic cavity and manual refinement.

DCE data were analyzed using model-free deconvolution\textsuperscript{19,20} implemented in a self-developed MatLab script (MathWorks, Natick, MA), yielding maps for the parameters: blood volume, blood flow, and mean transit time. The arterial input function was determined by drawing a region of interest in the pulmonary artery. Large vessels were removed using cross-correlation analysis.\textsuperscript{21}

For \textsuperscript{129}Xe CSSR measurements, Patz et al\textsuperscript{11} developed a model function of gas uptake to the dissolved phase as a function of CSSR delay time. This function with the free parameters septal wall thickness, surface-to-volume ratio, and capillary transit time was fitted to the uptake curves of the TP peak area to obtain whole-lung estimates of these parameters. Lineshape analysis of the high-resolution \textsuperscript{129}Xe dissolved-phase spectra was performed by fitting two complex Lorentzians in the frequency domain, yielding chemical shifts and T\textsubscript{2}* relaxation times.

### Statistical Analysis

Statistical tests were performed using MatLab R2014b (MathWorks, Ismaning, Germany). For comparison of data after challenge with baseline, the Wilcoxon signed-rank test was used ($\alpha = 0.05$ two-sided). Correlations of results from MRI with results from sputum analysis were assessed using Pearson’s correlation coefficient and significance of the correlation was assessed by a permutation test. Since this was an exploratory pilot study, no sample size justification is given.

### Results

Subject demographics and baseline clinical characteristics are shown in Table 1. Due to erroneous calibration of the MRI transmit system, the dissolved-phase imaging and high-resolution spectroscopy data after LPS inhalation in one subject contained only noise. The same subject did not produce sufficient amounts of sputum for assessment of cell counts.

Exemplary MR images before and after LPS challenge are shown in Fig. 1 and results from all MR measurements are summarized in Supporting Table 1. No clear changes of gas distribution were observed in \textsuperscript{129}Xe ventilation imaging and the ventilation defect percentage was not changed significantly, $P = 0.16$. 

### TABLE 1. Subject Demographics and Baseline Clinical Characteristics

| N   | 10 |
| Age (years) | 36 (31–46) |
| Sex | 7 male, 3 female |
| Height (cm) | 182 (176–183) |
| Weight (kg) | 79 (66–65) |
| Ethnicity | Caucasian |
| Hematocrit (%) | 42 (40–44) |
| Platelets ($10^9$/L) | 221 (198–245) |
| Neutrophils ($10^9$/L) | 2.8 (2.1–4.5) |
| Lymphocytes ($10^9$/L) | 1.6 (1.2–1.7) |
| Monocytes ($10^9$/L) | 0.4 (0.4–0.5) |
| Eosinophils ($10^9$/L) | 0.1 (0.1–0.2) |
| Creatinine (mg/dL) | 0.89 (0.83–0.99) |
| Bilirubin (mg/dL) | 0.61 (0.55–0.80) |
| Aspartate transaminase (U/L) | 26 (22–29) |
| Alanine transaminase (U/L) | 18 (17–24) |
| Glucose (mg/dL) | 83 (71–91) |
| Immunoglobulin E (IU/mL) | 33 (10–55) |

Values are median (25\textsuperscript{th}–75\textsuperscript{th} percentile) unless stated otherwise.
Both FEV₁ and FVC as percent of predicted value were significantly reduced after inhaled LPS challenge (Table 2). Cell counts from induced sputum are summarized in Supporting Table 2. Percentages of macrophages, neutrophils, and monocytes in sputum were significantly different compared to baseline, \( P = 0.002 \), while this was not the case for the percentage of lymphocytes.

There was a marked increase in neutrophil percentage in sputum after LPS inhalation (Fig. 2a). A significant reduction of the group median of whole-lung RBC-TP was observed in \(^{129}\text{Xe}\) dissolved-phase imaging after inhaled LPS challenge, 0.31, compared to baseline, 0.40, \( P = 0.004 \) (Fig. 2b). There also was a significant reduction in whole-lung RBC-GP in dissolved-phase imaging from 0.46 to 0.42, \( P = 0.020 \), and a strong trend for elevated TP-GP, \( P = 0.074 \). Figure 3 summarizes the results of CSSR spectroscopy before and after inhaled LPS challenge. The capillary transit time was significantly increased after inhalation of LPS, 2.48 sec, compared to baseline, 1.96 sec, \( P = 0.020 \). Exemplary \(^{129}\text{Xe}\) dissolved-phase spectra for the whole lung are depicted in Fig. 4a. Lineshape analysis exhibited a significant increase in \( T_{2}^* \) relaxation time of the TP phase after inhalation of LPS compared to baseline (Fig. 4b). The chemical shift of the RBC resonance was significantly reduced compared to baseline (Fig. 4c).

\(^{1}\text{H} \ T_{1} \) was significantly elevated after LPS challenge, 1187.8 msec, compared to baseline, 1157.6 msec, \( P = 0.027 \) (Fig. 2c). No significant changes of pulmonary blood volume, \( P = 0.64 \), pulmonary blood flow, \( P = 0.17 \), and mean transit time, \( P = 0.11 \), were observed in DCE MRI.

A significant correlation between the change of RBC-TP and change of neutrophil fraction from sputum analysis was observed (Fig. 5).

### TABLE 2. Results From Pulmonary Function Tests

|      | FEV₁ (% of predicted value) | FVC (% of predicted value) | FEV₁/FVC (%) |
|------|-----------------------------|-----------------------------|--------------|
| Baseline | 99.5 (95.7–113.8)          | 110.7 (97.9–112.9)          | 76.3 (75.2–78.5) |
| LPS    | 98.6 (86.1–107.1)          | 103.3 (92.1–110.1)          | 76.4 (74.0–80.5) |
| \( P \) | 0.037                      | 0.004                       | 0.842         |

Values are median (25th–75th percentile). FEV₁, forced expiratory volume in one second; FVC, forced vital capacity; LPS, lipopolysaccharide.
The feasibility of $^{129}$Xe MRI, $^1$H $T_1$ mapping and DCE MRI was investigated for noninvasive monitoring of the response of human lungs to inhaled LPS challenge as well as their relation to cell counts as established markers of inflammation. The results of our study show the feasibility and added value of MRI through noninvasive assessment of the inflammatory response at the alveolar level when compared to induced sputum collection.

While the RBC-TP ratio is sensitive for gas diffusion limitation,22 it is clear that there is also a contribution from perfusion to this quantity.23 The results of DCE imaging

FIGURE 2: (a) Significantly increased neutrophil percentage in sputum after inhaled LPS challenge, $P = 0.002$. (b) Whole lung mean RBC-TP is significantly reduced after inhaled LPS challenge, $P = 0.004$. (c) A significant increase in $T_1$ is observed after inhaled LPS challenge, $P = 0.027$. LPS, lipopolysaccharide; RBC, red blood cell; TP, tissue/plasma.

FIGURE 3: (a) CSSR gas uptake measurements at baseline and after inhaled LPS challenge. Surface-to-volume ratio is proportional to initial slope for low delay times, septal wall thickness is given by time to saturation and the slope for high delay times increases with blood flow velocity, ie, inverse of the capillary transit time. The level of saturation is proportional to the volume ratio of parenchyma to air spaces. (b) No significant change was observed for the CSSR-derived septal wall thickness parameter after LPS inhalation, $P = 0.193$. (c) There was a trend to an increased surface-to-volume ratio after challenge, $P = 0.084$. (d) A significantly increased ($P = 0.020$) capillary transit time corresponding to reduced blood flow velocity is observed after inhaled LPS challenge. There is one outlying data point with a very high capillary transit time of 65.6 sec after LPS. Without this subject, there still was a significant increase, $P = 0.040$. CSSR, chemical shift saturation recovery; LPS, lipopolysaccharide; S/V, surface-to-volume ratio; TP, tissue/plasma.
showed no significant changes in pulmonary parenchymal perfusion and pulmonary blood volume after LPS challenge. This suggests that the reduction of RBC-TP after inhaled LPS challenge is primarily attributable to pulmonary edema and not due to parenchymal hypoperfusion. The increase in proton $T_1$ is consistent with this interpretation, since water content increases $T_1$.8,24,25

The increased capillary transit time parameter from $^{129}$Xe CSSR measurements after inhaled LPS challenge is likely due to vasodilation, leading to reduced blood flow velocity at the capillary level. The fact that no such change is observed for mean transit time from DCE MRI suggests that hyperpolarized $^{129}$Xe MR is more specifically probing blood flow at the level of capillaries.

The increase of TP $T_2^*$ may suggest a reduced local heterogeneity of magnetic flux density due to reduced air-/alveolar surface interfaces due to septic edema and increased fluid content of the alveoli after LPS inhalation26 or a slower chemical exchange between TP and RBC phases. A similar increase of TP $T_2^*$ has previously been observed in idiopathic pulmonary fibrosis.22 It is also known that $^{129}$Xe chemical shift in RBCs is a marker for blood oxygenation27 and the reduction in chemical shift hence may suggest reduced oxygenation due to an oxygen diffusion limitation. Further research is necessary in order to validate this finding.

Septal thickening and a decreased septal surface density have been reported using histology and septic thickening has also been observed previously using CSSR in mouse models of inflammatory lung injury using LPS.28,29 One would have
expected the CSSR septal wall thickness and potentially also surface-to-volume ratio parameter to significantly increase after inhalation of LPS, which was not the case. This might have to do with the relatively low dose of inhaled LPS and small sample size in our study. In a previous reproducibility study, the mean coefficient of variation was found to be 4.7% for the wall thickness parameter and 12.2% for the surface-to-volume ratio. Another reason might be the dependence of these two quantities on lung inflation, which makes the method prone to errors despite the standardization of breathing maneuver.

It would have been possible in principle to perform this feasibility study in large animals. We decided, however, to perform it in healthy human volunteers since the translation of some results would not have been clear, for example because the spectral properties and even number of resonances of the hyperpolarized $^{129}$Xe dissolved phase are different among different species.

Recently, Svenningsen et al. found the volume of ventilation defects in hyperpolarized gas MRI after application of a bronchodilator to be quantitatively associated with the amount of eosinophils in sputum in severe asthmatics and thus suggested ventilation imaging for assessment of the inflammatory component of airway disease. Ventilation defects have also previously been observed using $^3$He MRI in mice after inhalation of a higher dose of 4–5 μg LPS. In comparison, our results indicate that in the presented human low-dose LPS inhalation model there are no clear changes in ventilation distribution, although the nonsignificant increase of group median ventilation defect percentage by 1.5% after LPS may point to a small change below our detection limit.

The correlation of change in RBC-TP and change in neutrophil fraction in induced sputum constitutes initial evidence that MRI is able to quantify the degree of inflammation in terms of cell counts. This relation still needs to be confirmed in future studies with larger cohorts.

One of the limitations of this study is the relatively small sample size. This makes the assessment of relationships between quantities derived from MRI and physiologic processes derived from cell counts difficult. Previous studies with positron emission tomography were able to show correlations of [$^{18}$F]fluorodeoxyglucose uptake and neutrophil counts from bronchoalveolar lavage in various diseases, although also only a weak correlation with $r^2 = 0.21$ was observed in a segmental LPS model of lung inflammation. Pulmonary perfusion measurements using DCE MRI also have various limitations with regard to physiological variations like the dependence on heart rate and inspiratory level as well as technical limitations. A limitation of our spectroscopic line-shape analysis is the negligence of non-Lorentzian effects in spectral broadening.

In conclusion, hyperpolarized $^{129}$Xe dissolved-phase MRI provides promising biomarkers, especially the RBC-TP ratio, for assessment of lung function at the alveolar level, which enable longitudinal monitoring of the response of human lungs to LPS challenge. $^{129}$Xe MRI and $^1$H T$_1$ mapping yield important complementary regional information in addition to the cell count derived from induced sputum and show promise as tools for monitoring inflammation in drug development.

**Acknowledgment**

Contract grant sponsor: German Center for Lung Research (DZL). Lipopolysaccharide (Clinical Center Reference Endotoxin) was kindly provided by Dr. Anthony Suffredini, National Institutes of Health, Bethesda, Maryland, USA.

**References**

1. Wang Y, Xu J, Meng Y, Adcock IM, Yao X. Role of inflammatory cells in airway remodeling in COPD. Int J COPD 2018;13:3341–3348.
2. Holz O, Tan L, Schaumann F, et al. Inter- and intrasubject variability of the inflammatory response to segmental endotoxin challenge in healthy volunteers. Pulm Pharmacol Ther 2015;35:50–59.
3. Hohlfeld JM, Schoenefeld K, Lavae-Mokhtari M, et al. Rufomustat attenuates pulmonary inflammation upon segmental endotoxin challenge in healthy subjects: A randomized placebo-controlled trial. Pulm Pharmacol Ther 2008;21:616–623.
4. Singh D, Siew L, Christensen J, et al. Oral and inhaled p38 MAPK inhibitors. Effects on inhaled LPS challenge in healthy subjects. Eur J Clin Pharmacol 2015;71:1175–1184.
5. Kips JC, Inman MD, Jayaram L, et al. The use of induced sputum in clinical trials. Eur Respir J 2002;20:47–50s.
6. Nightingale JA, Rogers DF, Barnes PJ. Effect of repeated sputum induction on cell counts in normal volunteers. Thorax 1998;53:87–90.
7. Vogel-Claussen J, Renne J, Hinrichs J, et al. Quantification of pulmonary inflammation after segmental allergen challenge using turbo-inversion recovery-magnitude magnetic resonance imaging. Am J Respir Crit Care Med 2014;189:650–657.
8. Renne J, Hinrichs J, Schönfeld C, et al. Noninvasive quantification of airway inflammation following segmental allergen challenge with functional MR imaging: A proof of concept study. Radiology 2015;274:267–274.
9. Jakob PM, Hillenbrand CM, Wang T, Schultz G, Hahn D, Haase A. Rapid quantitative lung $^1$H T1 mapping. J Magn Reson Imaging 2001;14:795–799.
10. Kern AL, Vogel-Claussen J. Hyperpolarized gas MRI in pulmonology. Br J Radiol 2018;91:20170647.
11. Patz S, Muradyan I, Hrovat MI, et al. Diffusion of hyperpolarized 129 Xe in the lung: A simplified model of 129 Xe septal uptake and experimental results. New J Phys 2011;13:015009.
12. Janssen O, Schaumann F, Holz O, et al. Lipopolysaccharide (Clinical Center Reference Endotoxin) was kindly provided by Dr. Anthony Suffredini, National Institutes of Health, Bethesda, Maryland, USA.
13. Möller W, Heimbeck I, Hofer TPJ, et al. Differential inflammatory response to inhaled lipopolysaccharide targeted either to the airways or the alveoli in man. PLoS One 2012;7:1–9.
14. Zhang J, Ruan W, Han Y, Sun X, Ye C, Zhou X. Fast determination of flip angle and T1 in hyperpolarized gas MRI during a single breath-hold. Sci Rep 2016;6:25854.
15. Qing K, Ruppert K, Jiang Y, et al. Regional mapping of gas uptake by blood and tissue in the human lung using hyperpolarized xenon-129 MRI. J Magn Reson Imaging 2014;39:346–359.
16. Kern AL, Gutberlet M, Qing K, et al. Regional investigation of lung function and microstructure parameters by localized 129 Xe chemical shift saturation recovery and dissolved-phase imaging: A reproducibility study. Magn Reson Med 2019;81:13–24.

17. Laub G, Kroecker R, syngo TWIST for dynamic time-resolved MR angiography. MAGNETOM Flash 2006;34:92–95.

18. He M, Kaushik SS, Robertson SH, et al. Extending semiautomatic ventilation defect analysis for hyperpolarized 129Xe ventilation MRI. Acad Radiol. 2014;21:1530–1541.

19. Östergaard L, Weisskoff RM, Chesler DA, et al. UMMPerfusion: An open source software tool towards quantitative MRI perfusion analysis in clinical routine. J Digit Imaging 2013;26:344–352.

20. Santini F, Patil S, Meckel S, Scheffler K, Wetzel SG. Double-reference cross-correlation algorithm for separation of the arteries and veins from 3D MRA time series. J Magn Reson Imaging 2008;28:646–654.

21. Kaushik SS, Freeman MS, Yoon SW, et al. Measuring diffusion limitation with a perfusion-limited gas—Hyperpolarized 129Xe gas-transfer spectroscopy in patients with idiopathic pulmonary fibrosis. J Appl Physiol 2014;117:577–585.

22. Phillips DM, Allen PS, Man SF. Assessment of temporal changes in pulmonary edema with NMR imaging. J Appl Physiol 1989;66:1197–1208.

23. Qiu J, Liu Y, Meckel S, Wetzel SG. Double-reference cross-correlation algorithm for separation of the arteries and veins from 3D MRA time series. J Magn Reson Imaging 2008;28:646–654.

24. Phillips DM, Allen PS, Man SF. Assessment of temporal changes in pulmonary edema with NMR imaging. J Appl Physiol 1989;66:1197–1208.

25. Kveder M, Zupanec I, Lahajnar G, et al. Water proton NMR relaxation spectroscopy in patients with idiopathic pulmonary fibrosis. J Magn Reson Med 1988;7:432–441.

26. Christman RA, Ailion DC, Case TA, et al. Comparison of calculated and experimental NMR spectral broadening for lung tissue. Magn Reson Med 1996;35:6–13.

27. Wolber J, Cherubini A, Leach MO, Bidone A. Hyperpolarized 129Xe NMR as a probe for blood oxygenation. Magn Reson Med 2000;43:491–496.

28. Månsson S, Wolber J, Drieuhaus B, Wolmer P, Golman K. Characterization of diffusion capacity and perfusion of the rat lung in a lipopolysaccharide disease model using hyperpolarized 129Xe. Magn Reson Med 2003;50:1170–1179.

29. De Souza Xavier Costa N, Ribeiro Junior G, Dos Santos Alemany AA, et al. Early and late pulmonary effects of nebulized LPS in mice: An acute lung injury model. PLoS One 2017;12:1–16.

30. Stewart NJ, Horn FC, Norquay G, et al. Reproducibility of quantitative indices of lung function and microstructure from 129 Xe chemical shift saturation recovery (CSSR) MR spectroscopy. Magn Reson Med 2017;77:2107–2113.

31. Pata S, Muradian I, Hrovat MI, et al. Human pulmonary imaging and spectroscopy with hyperpolarized 129Xe at 0.2T. Acad Radiol 2008;15:713–727.

32. Qing K, Ruppert K. Hyperpolarized xenon-129 dissolved–phase magnetic resonance imaging. In: Albert M, Hane F, eds. Hyperpolarized and inert gas MRI. From technology to application in research and medicine. Amsterdam: Elsevier; 2017:172.

33. Svenningsen S, Eddy R, Lim HF, Cox PG, Nair P, Parraga G. Sputum eosinophilia and magnetic resonance imaging ventilation heterogeneity in severe asthma. Am J Respir Crit Care Med 2018;197:876–884.

34. Olsson LE, Smailagic A, Önnervik PO, Hockings PD. 1H and hyperpolarized 3He MR imaging of mouse with LPS-induced inflammation. J Magn Reson Imaging 2009;29:977–981.

35. Chen DL, Ferkol TW, Mintun MA, Pittman JE, Rosenbluth DB, Schuster DP. Quantifying pulmonary inflammation in cystic fibrosis with positron emission tomography. Am J Respir Crit Care Med 2006;173:1363–1369.

36. Jones HA, Marino PS, Shakur BH, Morrell NW. In vivo assessment of lung inflammatory cell activity in patients with COPD and asthma. Eur Respir J 2003;21:567–573.

37. Chen DL, Rosenbluth DB, Mintun MA, Schuster DP. FDG-PET imaging of pulmonary inflammation in healthy volunteers after airway instillation of endotoxin. J Appl Physiol 2006;100:1602–1609.

38. Ley-Zaporozhan J, Molinari F, Risse F, Puderbach M, Schenk J-P, Kopp-Schneider A. Repeatability and reproducibility of quantitative whole-lung perfusion magnetic resonance imaging. J Thorac Imaging 2011;26:230–239.

39. Sourbron S. Technical aspects of MR perfusion. Eur J Radiol 2010;76:304–313.

40. Bier EA, Robertson SH, Schrank GM, et al. A protocol for quantifying cardiogenic oscillations in dynamic 129 Xe gas exchange spectroscopy: The effects of idiopathic pulmonary fibrosis. NMR Biomed 2019;32:e4029.