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Advance in Knowledge:

- Treatment Response Mapping (TRM) is a novel technique for quantitative assessment of regional lung ventilation changes in response to treatment.
- The net TRM integrated over the whole lungs ($\Delta R_{\text{net}}$) correlates with changes in spirometry (Pearson’s correlation: $\Delta FEV1$: $r = 0.70$, $\Delta FVC$: $r = 0.84$; $p<0.01$).
- In comparison to standard clinical outcome measures based on lung function, TRM adds information about the size and direction of the regional physiological response of the lungs.

Implication for Patient Care:

- Treatment Response Mapping (TRM) has potential for the assessment of regional lung interventions such as anti-inflammatory therapies or targeted therapies such as thermoplasty, endobronchial valve therapy and lung volume reduction surgery.
- Potential clinical applications for TRM are paediatric and longitudinal studies of lung disease progression as patients are not exposed to ionizing radiation.

Summary Statement:

Treatment Response Mapping (TRM) is introduced as a novel technique to provide regional quantitative information on changes in lung ventilation in response to therapy and is demonstrated in an asthma cohort by measuring bronchodilator response.
Abstract (248 words)

**Purpose:** To assess the magnitude of regional response to respiratory therapeutics in the lungs using Treatment Response Mapping (TRM) with hyperpolarized gas MRI. TRM is used to quantify regional physiological response in asthmatic adults using a bronchodilator challenge.

**Methods:** The study was approved by the national research ethics committee and performed with informed consent. Imaging was performed in 20 adult asthmatic patients using hyperpolarized $^3$He ventilation MRI. Two sets of baseline images were acquired before inhalation of a bronchodilator (Inhaled Salbutamol 400 mcg) and one set was acquired after. All images were registered for voxelwise comparison. Regional treatment response, $\Delta R(r)$, is calculated as the difference in regional gas distribution ($R(r) = \text{ratio of inhaled gas to total volume of a voxel when normalized for lung inflation volume}$) before and after intervention. A voxelwise activation threshold from the variability of the baseline images was applied to $\Delta R(r)$ maps. The summed global TRM ($\Delta R_{\text{net}}$) was then used as global lung index for comparison with metrics of bronchodilator response measured using spirometry and the global imaging metric, percentage ventilated volume (%VV).

**Results:** $\Delta R_{\text{net}}$ showed significant correlation ($p<0.01$) with changes in FEV$_1$ ($r=0.70$), FVC ($r=0.84$) and %VV ($r=0.56$). A significant ($p<0.01$) positive treatment effect was detected by all metrics, however $\Delta R_{\text{net}}$ showed a lower inter-subject coefficient of variation (CV=64%) than all of the other tests (CV≥99%).

**Conclusions:** TRM provides regional quantitative information on changes in inhaled gas ventilation in response to therapy. This method could be used as sensitive regional outcome metric of novel respiratory interventions.

Online supplemental material is available for this article.
INTRODUCTION

Lung function tests such as spirometry are widely used to clinically assess airflow obstruction and its reversibility and indices such as forced expiratory volume in 1 second (FEV<sub>1</sub>) are commonly accepted outcome measures in the assessment of therapies for obstructive lung disease. These techniques, whilst established in respiratory medicine, assess the lungs as one unit with limited sensitivity to regional ventilation changes (1). Imaging as a diagnostic tool can provide regional insight into alterations of both structure and function of the lungs and is increasingly being used as an outcome measure in the early phase evaluation of respiratory therapeutics (2). In particular, regionally specific therapies such as bronchial thermoplasty in asthma (3), endobronchial valve therapy (4) and lung volume reduction surgery in COPD (5) require regional information to assess the efficacy of intervention. Previous studies have used CT and computational fluid dynamics derived markers of airflow to assess functional changes after bronchodilator therapy (6). Nevertheless, those methods rely solely on models of ventilation inferred from structural CT images acquired at different levels of inspiration and repeated use of ionizing radiation which could be detrimental in particular in paediatric studies.

Functional lung imaging using hyperpolarized (HP) gas MRI provides images of lung ventilation in 3D in a short breath-hold. HP<sup>3</sup>He MRI has been shown to be a sensitive measure of ventilation heterogeneity in asthma (7, 8). While numerous metrics have been derived to describe ventilation heterogeneity from both HP<sup>129</sup>Xe and <sup>3</sup>He images using texture based methods like feature analysis or clustering methods such as k-means (9-12) efforts have been focused on cross-sectional assessment of cohorts. Longitudinal studies and therapy assessment have focused on the global outcome measure from HP<sup>3</sup>He images in particular percentage ventilated volume (%VV) or its counterpart the percentage defect volume (7, 13). While %VV has been shown to correlate with spirometry in asthma (14, 15), the method is limited by the binary classification of ventilated versus non-ventilated lung regions sacrificing much of the richness of the regional information on lung
ventilation heterogeneity present in the images. There is therefore a need for imaging metrics that fully explore the regional sensitivity of these high-resolution images of lung function to assess success of intervention.

Treatment Response Mapping (TRM) is introduced as a novel technique to provide regional quantitative information on changes in lung ventilation in response to therapy and is demonstrated in an asthma cohort by measuring bronchodilator response.

Material and Methods

The study was approved by the national research ethics committee, and written patient consent was obtained. All data were acquired between February 2012 and June 2013 and the study was funded in parts by Novartis. One author (R.K.) is an employee of Novartis. The authors not employed by Novartis had full control of the data and the information submitted for publication.

Study Population and Design

20 patients (10/10, female/male) diagnosed with moderate-to-severe asthma (Global Initiative for Asthma (GINA) step 2 – 5 (16)), were studied in this retrospective analysis. Patients age range was 21-73 years. Patients were tested for response to bronchodilator (400mcg Salbutamol) with HP gas ventilation MR imaging and with spirometry. Patient demographics and results from pulmonary function tests are shown in Table 1.

MRI Data Acquisition

Prior to the imaging acquisition visit, participants refrained from using any short acting bronchodilators for at least six hours. Patients were then imaged in a supine position on a clinical 1.5 T MRI scanner (GE HDx, Milwaukee, WI, USA) with a dedicated MR $^3$He RF coil. $^1$H MRI
was performed with the system’s body coil. Each image was acquired upon inhalation of 1 liter of
gas from a Tedlar bag (350 ml HP\(^3\)He mixed with 650 ml N\(_2\)) from functional residual capacity.
Prior to scanning, subjects were trained in the breathing manoeuvre. Three ventilation MRI scans
were performed in separate breath-holds; two at baseline within 5 minutes of each other, then an
additional scan 20 minutes after bronchodilator to assess short-term airway responsiveness (17)
(Figure 1). Each breath-hold scan consisted of: (1) functional images (HP\(^3\)He ventilation,
resolution=3x3x10mm, duration=9s) and (2) structural images (\(^1\)H anatomy,
resolution=3x6x10mm, duration=4s). To switch between \(^3\)He and \(^1\)H MR-imaging the scanner
required ~3-5s leading to a total breath-hold duration of under 18 seconds in all cases. Since both
functional and structural images are acquired back-to-back during a single breath-hold, they are
intrinsically co-registered (18).

Image processing algorithm for Treatment Response Mapping (TRM)

Structural proton images were first registered between the different time points and the resulting
transformation was applied to the same breath-hold \(^3\)He ventilation images (19). This avoided the
registration of the \(^3\)He ventilation images being influenced by regional changes of gas distribution
between scans (see online supplementary material). Then images were segmented to extract the
ventilated lung volume using ScanIP (Simpleware Ltd., Exeter, UK).

Regional ventilation was then calculated from image intensity for each dataset. Ventilation as
regional gas distribution in the lung is quantified by conversion of image intensity to the volume
fraction occupied by the inhaled tracer gas in each voxel, this requires knowledge of voxel volume
\((V_{\text{voxel}})\) and inhaled gas volume \((V_{\text{bag}})\) as described by Tzeng et al. (10):

\[
R'(r) = \frac{I(r)}{V_{\text{voxel}}} \cdot \frac{V_{\text{bag}}}{I_{\text{tot}}}
\]

Eq. 1
Where $I(r)$ is the regional image intensity at position $r(x,y,z)$, $I_{tot}$ the integrated image intensity of all ventilated areas in the image. $R'(r)$ is the resulting regional gas volume fraction in each voxel.

For example, in the trachea directly after inhalation the gas composition is usually the same as that inhaled from the bag and hence $R' = 1$.

In order to compare $R'(r)$ between datasets, the differences in gas dilution of the tracer gas in the ventilated airspaces have to be taken into account. Following intervention airway opening/closure will likely result in differences in ventilated lung volume. These variations can cause dilution of the tracer gas concentration when it is inhaled in the same dose from breath to breath. The ventilated lung volume after treatment ($V_{LTx}$) was chosen as the reference point and the normalized gas volume fraction $R$ was computed as follows:

$$R(r) = R'(r) \cdot \frac{V_{L}}{V_{LTx}}$$  \hspace{1cm} \text{Eq. 2}

Where $V_{L}$ is the ventilated lung volume calculated from the image of interest. Equation 2 assumes that regional gas concentration scales uniformly with lung volume changes across all voxels in the lungs.

The difference of gas volume fraction, $\Delta R(r)$, before and after intervention is then calculated for all positions ($r$) in order to quantify TRM. $\Delta R(r)$ is in effect the regionally measured redistribution of the gas mixture when inhaling identical doses before and after intervention. Like $R(r)$, the TRM, $\Delta R(r)$ is a gas volume fraction, which measures the size and direction of ventilation change due to intervention. The major airways were excluded, as they do not contribute to gas exchange and can be expected to be fully ventilated in each breath. To account for ventilation changes related to physiological baseline variability from scan-to-scan, a voxel-wise baseline variability map, $\Delta R_{B}(r)$, is calculated as the standard deviation of the differences in gas volume fraction $R(r)$ between the two (filtered) baseline scans. $\Delta R(r)$ is set to zero (no effect) for voxels whose $|\Delta R(r)| \leq |\Delta R_{B}(r)|$ and is displayed as white voxels. For remaining voxels, a positive $\Delta R(r)$ (improvement in local ventilation) is shown with a green color scale and a negative $\Delta R(r)$ (reduction in local ventilation)
with a red color scale. In addition, a global value for net treatment response over the whole lungs, \( \Delta R_{\text{net}} \), was calculated:

\[
\Delta R_{\text{net}} = \sum_r \Delta R(r) \cdot V_{\text{voxel}}
\]

Eq. 3

\( \Delta R_{\text{net}} \) is summed over the lung volume and can be expressed as the percentage (of the inhaled gas mix =1 liter) or alternatively in ml as the volume of the gas dose that is delivered to newly ventilated regions of the lung following inhalation of a 1 liter dose. We emphasise that expression of \( \Delta R_{\text{net}} \) in ml is not to be confused with the volume of the lungs that opens up in response to the bronchodilator, instead it is the amount of the tracer gas (from a 1 liter total inhaled dose), that reaches newly ventilated lungs. For an inhaled gas mix of 1 liter a theoretical redistribution of \( \Delta R_{\text{net}}=100\% \) or +1liter would therefore mean a complete redistribution of all inhaled gas in previously non- or little-ventilated regions. The metric quantifies the sum of regional gas redistribution as result of the same dose inhaled before and after an intervention. Accordingly, the global positive and negative change can be calculated by taking only positive or negative \( \Delta R(r) \) into account. An analysis of error propagation related to these steps of calculation of TRM is presented in the online supplement.

Percentage ventilated volume

\( \%VV \) was calculated as the ratio of lung ventilated volume (from segmented \(^3\)He ventilation images) to the total volume of the lung in the thorax (from segmentation of the \(^1\)H image) as described previously (18).

Spirometry

Spirometry was performed with a rolling seal Vitalograph spirometer (Vitalograph, Buckingham, UK) before and 20 minutes after bronchodilator inhalation according to ATS/ERS guidelines (20). A minimum of three acceptable FVC manoeuvres were performed and the highest FEV1 and FVC
of the acceptable curves were recorded. Percent predicted values were calculated as described in (21). All pre-bronchodilator tests were performed after all bronchodilators had been withheld for at least six hours.

Statistics

Statistical analysis was performed using GraphPad Prism (GraphPad Software Inc., San Diego, CA, USA). All datasets were first tested for normality using a D’Agostino-Pearson omnibus normality test. Significance of treatment effect was tested using a paired t-test or in the case of a non-normal distribution the Wilcoxon signed rank. The probability that the null hypothesis ($H_0 =$ no treatment effect is found) can be rejected was tested (for $p<0.05 H_0$ rejected). Values are reported as median ± interquartile range (25th-75th quartile). Correlations were calculated using Pearson’s $r$ correlation (Spearman’s correlation in case of non-normality) and the correlation coefficient and significance level of correlation ($p$-value) are reported.
RESULTS

An overview of results from the global MRI metrics ($\Delta R_{\text{net}}$, %VV) and lung function test measurements (FEV$_1$, FVC) are presented in Table 2 (details of each patient in Table E2). TRM indicated an overall positive effect of the bronchodilator in agreement with other metrics and the p-values of paired t-tests are shown in Table 2. An average inter-subject baseline variability $\Delta R_B$ of 4.74%±4.68% was found. Example $^3$He ventilation images and the resulting $\Delta R$ maps from Patient 2 are shown in Figure 2. 3D renders of TRM from 4 more patients are shown in Figure 3. Figure 4 shows correlation of $\Delta R_{\text{net}}$ with changes in the other metrics evaluated: %VV $r = 0.56$ (p = 0.01), FEV$_1$ $r = 0.70$ (p < 0.01) and FVC $r = 0.84$ (p<0.01). $\Delta R_{\text{net}}$ and FEV$_1$/FVC did not correlate significantly. The inter-subject coefficient of variation (CV) of $\Delta R_{\text{net}}$ was 64% which is much lower than the CV of %VV and spirometry (both ≥99%). Figure 5 shows the changes as assessed by all methods for each individual subject and their collective median (interquartile range).
DISCUSSION

Treatment response mapping (TRM) of changes in lung ventilation in response to therapy is demonstrated here with pre and post bronchodilator imaging in a cohort of asthma patients. The effects of bronchodilator are typically clinically assessed with spirometry, airway resistance or static lung volume measurement. While being quick and easy to repeat, regional changes in lung ventilation and airway opening/closing are not accessible with these methods. This is likely to reduce the sensitivity of these clinical tests to subtle but still clinically significant changes in ventilation in particular when the treatment is focused on a small lung region (e.g. bronchothermoplasty or endobronchial valves). Imaging as a diagnostic tool can provide regional insight into alterations of both structure and function of the lung. Airway wall measurements and models of lobar ventilation change from inspiratory and expiratory computed tomography (CT) have both been proposed as image based outcome measures in asthma (6), but the repeated exposure to ionizing radiation is an issue. HP gas MRI directly measures functional information from the lungs without ionizing radiation and TRM is a potentially powerful tool for longitudinal and paediatric studies. Previously, oxygen enhanced MRI has been used to measure changes induced by bronchodilator and corticosteroids (22). However, it is unclear to what extent this represents changes in ventilation as signal changes are caused by interaction of protons with $O_2$ dissolved in tissue and blood (23).

Signal intensity from hyperpolarized gas MR images is proportional to the distribution of inhaled tracer gas in the lung and is therefore a direct measure of ventilation. The TRM method demonstrated here uses this relation to regionally quantify the magnitude of ventilation changes. The inherent baseline variability of the images from scan-to-scan without any intervention is also taken in to account in the algorithm and defines a voxel-wise threshold for ‘treatment response’.
This is important as it has been previously shown that originally non-ventilated regions of the lung can change size and position on the same day in asthma patients without intervention (13).

One of the most striking findings was the close correlation of FVC and FEV$_1$ with global TRM metric $\Delta R_{\text{net}}$. The fact that imaging and spirometry were not performed on the same day represents a limitation of this study and might explain why the correlations between spirometry and $\Delta R_{\text{net}}$ are not stronger. Although all of the tested metrics showed a significant effect of the bronchodilator ($p<0.01$), the largest changes were found from $\Delta R_{\text{net}}$ (Figure 5). The inter-subject coefficient of variation of $\Delta R_{\text{net}}$ was also smaller (64%) than for the other metrics ($\geq 99\%$).

Some improvements in FVC were high (39% for Patient 2) and might reflect recruitment of air spaces due to decreased gas trapping, an assumption that is supported by increases in $\%VV$ (5.8% for Patient 2) in most patients. The weaker correlation ($r = 0.56$) of $\Delta R_{\text{net}}$ with changes in $\%VV$ supports the assumption, that changes in ventilation are not only a result of a net increase in viable ventilated airspace volume, but also reflect changes in heterogeneity of the magnitude of the ventilation which is regionally elucidated with the TRM method.

In patients with a limited spirometric response (<12%) to bronchodilator, TRM effectively distinguishes regions of the lung with an increase in ventilation from regions demonstrating reduced ventilation. This lends some regional evidence to the possibility that bronchodilators are not always effective and can be potentially detrimental in selected regions of the lung in asthma. Similar observations have been reported in other imaging biomarker studies (22). Nevertheless, negative $\Delta R(r)$ can result from two other mechanisms; while a degradation of ventilation is observed in practice in some lung regions after treatment, other regions of negative $\Delta R(r)$ might result from initially hyperventilated regions of lung that were perhaps compensating for obstruction elsewhere, then returning to more even levels after bronchodilator application. Understanding of physiological
mechanism and phenotyping these ‘red regions’ of worsened ventilation warrants further investigation.

In conclusion, TRM is able to complement the current techniques for regionally quantifying changes in ventilation in the lungs. The future clinical potential of the method lies in determining the regional response of the lungs to new therapies where established lung function tests do not provide sensitive enough outcome measures. This sensitivity to local changes in ventilation could also result in a reduction of patient cohort numbers required to confirm success of a treatment in a clinical trial.

Current limitations of the technique are the prolonged breath-hold required for image acquisition and the requirement for HP gas MR-imaging infrastructure for effective delivery in a clinical setting. Recent advances may help overcome this barrier in years to come as the technique may also be directly applicable to ventilation images acquired using other gases to image lung function such as HP $^{129}$Xe or $^{19}$F fluorinated gases like Perfluoropropane (24, 25).
References


Table 1. Overview of demographics and results from pulmonary function tests in 20 asthma patients. All values presented as mean (± standard deviation), measured before application of the bronchodilator. Spirometry was performed with a rolling seal Vitalograph spirometer (Vitalograph, Buckingham, UK) according to ERS/ATS guidelines (20).

<table>
<thead>
<tr>
<th>Study participants (n=20)</th>
</tr>
</thead>
</table>
| Age (years)              | 51±12  
| Sex (female/male)        | 10/10  
| Height (m)               | 1.66±0.07  
| Weight (kg)              | 82±15  
| BMI (kg/m²)              | 30±5  
| GINA                     | 4.10±0.68  
| Pack years               | 0.62±1.91  
| RV (L)                   | 2.78±1.18  
| TLC (L)                  | 6.23±1.54  
| FEV₁(%pred), mean±SD (range) | 71±28, (27-122)  
| FVC(%pred), mean±SD (range) | 92±22, (54-138)  
| FEV₁/FVC(%pred), mean±SD (range) | 75±17, (31-83)  

BMI = body mass index, %pred = percent predicted, GINA = global initiative for asthma classification (16), pack years = lifetime tobacco exposure, 1 pack year defined as 20 cigarettes a day for a year; RV = residual volume, TLC = total lung capacity, FEV₁ = forced expiratory volume in 1 second; FVC = Forced vital capacity. Predicted values calculated using GLI equations (21).
Table 2. Summary of outcome measures used to assess treatment response including the net treatment effect ($\Delta R_{\text{net}}$). All values expressed as median (interquartile range: 25th-75th quartile).

Patient specific values are shown in Table E2 (online) alongside scatter plots in Figure E1.
<table>
<thead>
<tr>
<th>Outcome metric</th>
<th>Treatment: Bronchodilator (BD)</th>
<th>Absolute measurement</th>
<th>postBD</th>
<th>preBD</th>
<th>paired t-test</th>
<th>p-value</th>
<th>Difference (postBD-preBD)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>preBD</td>
<td>postBD</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Absolute measurement</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>FEV&lt;sub&gt;1&lt;/sub&gt; (%pred)</td>
<td>72(44-96)</td>
<td>79(58-100)</td>
<td>0.0002</td>
<td>6.3(4.5-13)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>FVC (%pred)</td>
<td>85(76-110)</td>
<td>93(85-120)</td>
<td>0.004</td>
<td>5.2(1.9-8.3)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>FEV&lt;sub&gt;1&lt;/sub&gt;/FVC(%)</td>
<td>68(48-73)</td>
<td>88(65-91)</td>
<td>&lt;0.0001</td>
<td>3.6(2.4-5.5)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>%VV (imaging)</td>
<td>88(83-94)</td>
<td>93(90-97)</td>
<td>0.0009§</td>
<td>3.5(0.2-5.7)</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Differential measure</th>
<th>positive ΔR&lt;sub&gt;net&lt;/sub&gt;</th>
<th>negative ΔR&lt;sub&gt;net&lt;/sub&gt;</th>
<th>paired t-test</th>
<th>p-value</th>
<th>ΔR&lt;sub&gt;net&lt;/sub&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>ΔR&lt;sub&gt;net&lt;/sub&gt; (%)</td>
<td>20(16-26)</td>
<td>9(7-11)</td>
<td>&lt;0.0001§</td>
<td>11(6.6-14)</td>
<td></td>
</tr>
<tr>
<td>ΔR&lt;sub&gt;net&lt;/sub&gt; (ml)</td>
<td>210(160-260)</td>
<td>92(71-110)</td>
<td>&lt;0.0001§</td>
<td>110(66-140)</td>
<td></td>
</tr>
</tbody>
</table>

*CV = coefficient of variation as ratio of standard deviation to mean of treatment effect.

§ non-normally distributed data, Wilcoxon signed rank test was used. **Figure legends**

**Figure 1:** Image processing workflow from left to right: 3D image sets (proton and HP gas ventilation image within a single breath-hold) are acquired twice at baseline to calculate baseline variability. 20 minutes after bronchodilator application another 3D set is acquired. The difference between pre- and post-bronchodilator images are plotted as treatment response ΔR(r) maps and compared to a regional treatment activation threshold from baseline variability, ΔR<sub>th</sub>(r). The dataset is from a 54-year-old male, Patient 2.

**Figure 2:** Examples of coronal treatment response maps (top row), MR ventilation images at baseline (middle row) and post bronchodilator inhalation (bottom row), from Patient 2 (54 years,
male). The positive $\Delta R_{\text{net}} = 35.1\%$ is compared to the negative $\Delta R_{\text{net}} = 5.5\%$ resulting in a total $\Delta R_{\text{net}}$ of $+29.7\%$.

Figure 3: Example 3D rendered treatment response maps from 4 volunteers and the corresponding histograms.

**Figure 4:** Correlation of $\Delta R_{\text{net}}$ from histograms (in %) with changes in (a) percentage ventilation volume from before and after treatment ($\Delta \% \text{VV}$) with a Spearman’s $r = 0.56$, $P = 0.01$, (b) Correlation of $\Delta R_{\text{net}}$ with changes in FEV$_1$ from treatment ($\Delta \text{FEV}_1$) with a Pearson’s $r = 0.70$, $P = 0.004$ and (c) Correlation of $\Delta R_{\text{net}}$ with FVC with a Pearson’s $r = 0.84$, $P < 0.001$. Dotted lines show 95% confidence intervals.

**Figure 5:** Comparison of changes from bronchodilator visualizing the data from Table 2. Each plot shows median ± interquartile range interval including each individual data point. Changes are expressed as difference in percent predicted (FEV$_1$/FVC, FVC, FEV$_1$), difference in %VV before and after treatment and percent $\Delta R_{\text{net}}$. The plot shows that the treatment effect found from $\Delta R_{\text{net}}$ is greater on average and the standard deviation is smaller than those of spirometric indices.
Abbreviations used:

- %VV Percentage Ventilated Volume
- $\Delta R_B$ Baseline variability; change in ventilation fraction in each voxel between the two images acquired at baseline (before application of the bronchodilator)
- $\Delta R$ Treatment Response; change in ventilation fraction in each voxel between images acquired before and after application of the bronchodilator
- COPD Chronic obstructive pulmonary disease
- CT Computed tomography
- $\text{FEV}_1$ Forced expiratory volume in 1 second
- FVC Forced vital capacity
- HP Hyperpolarized
- MRI Magnetic resonance imaging
- $\Delta R_{\text{net}}$ Net treatment response over the whole lung as a result of repeated inhalation of 1 liter of the gas mix containing the hyperpolarized $^3$He gas; expressed in ml and as percent of the inhaled gas volume (1 liter)
- PFP Perfluoropropane gas is a fluorinated gas (used for lung imaging)
- TRM Treatment response map
- VH Ventilation heterogeneity
**Details of MR image acquisition**

Patients were imaged using a whole-body clinical 1.5 T MRI scanner (GE HDx, Milwaukee, WI, USA) with a quadrature transmit-receive vest coil tuned to 48.65MHz for $^3$He imaging (Clinical MR Solutions, Brookfield, WI, USA) and the quadrature transmit-receive body coil for proton imaging. Patients were trained in the breathing manoeuvre outside the MR scanner before scanning and once more inside the MRI scanner before administration of the HP $^3$He.

Helium was polarized on site to approximately 25% using a rubidium spin exchange polarizer (GE Healthcare, Amersham, UK). Patients inhaled 350 ml $^3$He mixed with 650 ml N$_2$ from a Tedlar bag (Jensen Inert Products, Coral Springs, Florida, USA) from functional residual capacity (FRC). Ventilation ($^3$He) and Anatomical ($^1$H) images were acquired back-to-back during one breath-hold covering the same volume (I).

Table E1. Summary of imaging parameters.

<table>
<thead>
<tr>
<th>Scan</th>
<th>MRI Sequence</th>
<th>Repetition time (ms)</th>
<th>Echo time (ms)</th>
<th>Flip angle (°)</th>
<th>Bandwidth (kHz)</th>
<th>Field of view (cm) frequency x phase</th>
<th>Matrix size frequency x phase</th>
<th>Pixel size (mm) frequency x phase</th>
<th>Slice thickness (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ventilation ($^3$He)</td>
<td>SPGR</td>
<td>3.6</td>
<td>1.1</td>
<td>8</td>
<td>62.5</td>
<td>38.4 x 30.7</td>
<td>128x102</td>
<td>3x3</td>
<td>10</td>
</tr>
<tr>
<td>Anatomical ($^1$H)</td>
<td>SSFP</td>
<td>2.4</td>
<td>2.4</td>
<td>50</td>
<td>167</td>
<td>38.4 x 38.4</td>
<td>128x64</td>
<td>3x6</td>
<td>10</td>
</tr>
</tbody>
</table>
Challenges in calculating treatment response maps

Quantifying regional ventilation changes between longitudinal time-points is a challenge in lung imaging due to patient re-positioning and differences in lung volume between acquisitions. The problem of lung volume variation between separate breaths has been highlighted previously (1) in the calculation of percent ventilated volume (%VV), where acquisition of proton images and ventilation images in separate breaths has been shown to considerably influence the reliability of %VV. Here %VV was calculated from ventilation and anatomical images acquired within the same breath to avoid this problem. When calculating treatment response maps, image registration with the same breath $^1$H anatomical images was used to overcome differences in lung volume of ventilation images acquired at different breath-holds and patient movement between time-points. This process reduces image registration errors introduced by morphological changes in gas distribution in the ventilation images that arise from response to treatment. Registration of images in this way then allows regional comparison of ventilation changes and treatment response mapping over multiple time points.

Images were acquired with the patient in a similar position within the coil and using the same sequence parameters each time, allowing a comparison of relative changes in image intensity. It is acknowledged that lung units with very low ventilation, resulting in a long time constant for gas wash-in, may not be captured with a single-breath ventilation imaging method (2).

Error in the calculation of ΔR

The quantification of treatment response is sensitive to the amount of HP gas inhaled. It is assumed that a total of 1 liter (350ml $^3$He and 650 N$_2$) is precisely dispensed and fully inhaled. Patients are trained in the inhalation process before entering the scanner to assure a successful emptying and
inhalation of the contents of the bag of gas. When dispensing the gas, a typical dispense accuracy of ±25ml in the volume of the bag can be achieved. The contribution of the inhaled bag volume to the calculation of the local voxelwise ventilation fraction can be derived from Equation 1 of the main text as follows:

$$R(\mathbf{r}) = \frac{I(\mathbf{r})}{V_{\text{voxel}}} \cdot \frac{V_{\text{bag}}}{V_{\text{tot}}}$$

Eq. E1

where $I(\mathbf{r})$ is the regional image intensity, $V_{\text{voxel}}$ is the volume of a voxel, $V_{\text{bag}}$ is the volume of the bag containing the HP $^3$He mixture (1 liter).

This allows a simple error estimation of how the bag volume $V_{\text{bag}}$ contributes to the error in voxelwise estimate of fractional ventilation:

$$\frac{\partial R}{R} = \sqrt{\left(\frac{\partial V_{\text{bag}}}{V_{\text{bag}}}\right)^2} = 2.5\%$$

Eq. E2

Thus this results in a propagated error in voxel fractional ventilation of less than 2.5%. As the treatment response measurement, $\Delta R(\mathbf{r})$ is the result of a subtraction of two ventilation fractions (R(\mathbf{r}) from two images) the maximum potential error is compounded to 3.5%.

The effect of image registration on $\Delta R(\mathbf{r})$ was not investigated. Nevertheless, the above discussion shows how effects were mitigated and it can therefore be assumed that effects from mis-registration are negligible.

Appendix Figures

Figure E1: Plots of all of the tested methods are shown. Individual plots show each data-point, median and interquartile range. (a) Net Treatment response ($\Delta R_{\text{net}}$), a significant response to
bronchodilator treatment was found. Although all methods show significant sensitivity to effects of bronchodilators, negative $\Delta R_{\text{net}}$ is tightly clustered around the mean value of 10% and both groups (positive and negative $\Delta R_{\text{net}}$) show visibly clearer separation. Positive treatment response (positive $\Delta R_{\text{net}} = 20\% \pm 7\%$) over the cohort was found to be significantly greater than average negative treatment response (negative $\Delta R_{\text{net}} = 10\% \pm 4\%$) ($P<0.0001$, CI of median = [8.8-12.4]). (b) %VV before and after bronchodilator application. Significant response to bronchodilator treatment was found ($p<0.01$). Percentage ventilated volume (%VV) also showed significant increase ($P<0.001$, CI of median = [0.41, 5.4]) after bronchodilator application. (c) FEV$_1$ before and after bronchodilator application, a significant response to bronchodilator treatment was found ($p<0.01$). FEV$_1$ also increased significantly ($P=0.0001$, CI of median = [5.1, 12.2]) in response to bronchodilator inhalation as well as FVC ($P=0.0001$, CI of median= [2.8, 8.0]). (d) FVC before and after bronchodilator application. Significant response to bronchodilator treatment was found ($p<0.01$).
Table E2: An overview of all outcome measure from each individual asthma patient imaged.

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<th>Post-Bronchodilator</th>
<th>Pre-PostBD FEV1</th>
<th>Treatment response mapping</th>
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