

Clinical correlates of lung ventilation defects in asthmatic children

Talissa A. Altes, MD,^a John P. Mugler III, PhD,^{b,c} Kai Ruppert, PhD,^d Nicholas J. Tustison, DSc,^b Joanne Gersbach, RN,^b Sylvia Szentpetery, MD,^e Craig H. Meyer, PhD,^{b,c} Eduard E. de Lange, MD,^b and W. Gerald Teague, MD^e *Columbia, Mo, Charlottesville, Va, and Cincinnati, Ohio*

Background: Lung ventilation defects identified by using hyperpolarized 3-helium gas (³He) lung magnetic resonance imaging (MRI) are prevalent in asthmatic patients, but the clinical importance of ventilation defects is poorly understood. **Objectives:** We sought to correlate the lung defect volume quantified by using ³He MRI with clinical features in children with mild and severe asthma.

Methods: Thirty-one children with asthma (median age, 10 years; age range, 3-17 years) underwent detailed characterization and ³He lung MRI. Quantification of the ³He signal defined ventilation defect and hypoventilated, ventilated, and well-ventilated volumes.

Results: The ventilation defect to total lung volume fraction ranged from 0.1% to 11.6%. Children with ventilation defect percentages in the upper tercile were more likely to have severe asthma than children in the lower terciles ($P = .005$). The ventilation defect percentage correlated ($P < .05$ for all) positively with the inhaled corticosteroid dose, total number of controller medications, and total blood eosinophil counts and negatively with the Asthma Control Test score, FEV₁ (percent predicted), FEV₁/forced vital capacity ratio (percent predicted), and forced expiratory flow rate from 25% to 75% of expired volume (percent predicted).

Conclusion: The lung defect volume percentage measured by using ³He MRI correlates with several clinical features of asthma, including severity, symptom score, medication

requirement, airway physiology, and atopic markers. (J Allergy Clin Immunol 2015;■■■:■■■-■■■.)

Key words: Hyperpolarized, ³He magnetic resonance imaging, ventilation defects, ventilation heterogeneity, severe asthma, childhood asthma

Lung ventilation defects are regions of absent hyperpolarized 3-helium gas (³He) signal identified by using hyperpolarized noble gas magnetic resonance imaging (MRI) in healthy volunteers and asthmatic patients.¹⁻³ Proposed mechanisms of ventilation defects vary from obstruction of the proximal airways to closure of acinar lung structures.⁴⁻⁶ Ventilation defects form more often in the lung bases in asthmatic patients and correspond spatially to regions of air trapping mapped by using multidetector computed tomography (CT).⁷ In repeat MRI studies of defects in asthmatic adults, approximately two thirds of ventilation defects do not change in size or location over time.⁸ Ventilation defects increase in size after methacholine-induced bronchospasm⁹ and decrease in size but do not completely resolve after inhalation of the bronchodilator albuterol.^{2,6}

Lung ventilation defects occur with increased prevalence in adults and children with asthma.^{10,11} Whereas much is known about the spatiotemporal features of lung ventilation defects in asthmatic patients, the clinical implications of ventilation defects are not well understood. In an original report the number of defects per image slice correlated with asthma severity and degree of airflow limitation.¹⁰ In a recent report the ventilation defect volume percentage correlated significantly with older age, lower FEV₁/forced vital capacity (FVC) ratio, higher expired nitric oxide level, greater methacholine responsiveness, and airway wall thickness estimated by using chest CT.¹² However, these studies did not include children, and important clinical features of asthma, including symptom control, treatment, and markers of inflammation, were not evaluated. This is an important gap insofar as even children with mild asthma and normal spirometric results can have ventilation heterogeneity and closure of the peripheral airways.¹³ Young children might be at increased risk for the formation of ventilation defects because of the small size of their airways and, in children before 2 years of age, the relative instability of the chest wall.¹⁴

Advances in rapid image sequencing methods have facilitated the acquisition of high-quality hyperpolarized gas magnetic resonance images in preschool children.¹⁵ Furthermore, improvements in image processing and signal intensity analysis can accurately measure lung volume compartments.¹⁶ We applied these innovations in a sample of asthmatic children to study whether the lung defect volume percentage, as measured by using hyperpolarized lung MRI, correlates with a range of clinical features. We hypothesized that the ventilation defect volume

From ^athe Department of Radiology, University of Missouri School of Medicine, Columbia; ^bthe Division of Medical Imaging Research, Department of Radiology and Medical Imaging, and ^cthe Child Health Research Center, Department of Pediatrics, University of Virginia School of Medicine, Charlottesville; ^dthe Department of Biomedical Engineering, University of Virginia, Charlottesville; and ^ethe Center for Pulmonary Imaging Research, Cincinnati Children's Hospital.

Supported by the National Institutes of Health/National Heart, Lung, and Blood Institute Severe Asthma Research Program (SARP), 1U01HL109250-1 (to W.G.T.), the Ivy Foundation (to W.G.T.), and the Hartwell Foundation (to T.A.A.).

Disclosure of potential conflict of interest: T. A. Altes has received a grant from the Hartwell Foundation. J. P. Mugler has consultant arrangements with and has received grants from Siemens Medical Solutions. K. Ruppert is employed by the University of Virginia and Cincinnati Children's Hospital. C. H. Meyer has received grants from the National Institutes of Health (NIH)/National Heart, Lung, and Blood Institute (NHLBI); the Ivy Foundation; the Hartwell Foundation; and Siemens and has a patent through the University of Virginia. W. G. Teague has received a grant from the NIH/NHLBI and TEVA and has received payment for lectures from Genentech, Merck, and TEVA. The rest of the authors declare that they have no relevant conflicts of interest. Received for publication November 3, 2014; revised August 18, 2015; accepted for publication August 21, 2015.

Corresponding author: W. Gerald Teague, MD, Department of Pediatrics, Division of Respiratory Medicine, Allergy, and Immunology, Child Health Research Center, University of Virginia School of Medicine, PO Box 30086, Charlottesville, VA 22908. E-mail: wgt2p@virginia.edu.

0091-6749/\$36.00

© 2015 American Academy of Allergy, Asthma & Immunology
<http://dx.doi.org/10.1016/j.jaci.2015.08.045>

Abbreviations used

ATS:	American Thoracic Society
CT:	Computed tomography
FEF ₂₅₋₇₅ :	Forced expiratory flow rate from 25% to 75% of expired volume
FVC:	Forced vital capacity
³ He:	Hyperpolarized 3-helium gas
MRI:	Magnetic resonance imaging
RF:	Radio frequency

percentage would be higher in children with severe asthma and correlate not only with the degree of airflow limitation but also indicators of asthma control, treatment, and inflammation.

METHODS

The study sample included children with a physician's diagnosis of asthma referred for evaluation to a regional medical center. The study was approved by the Institutional Review Board of the University of Virginia Health System (#157200). ³He gas was administered under IND 57866 held by Dr E. de Lange. The parents of enrollees provided informed consent, and children of appropriate age provided assent.

Asthma confirmation and characterization procedures

To fit the inclusion criteria, enrollees were 3 to 17 years of age, mature enough to cooperate with the MRI procedures, and had current asthma based on a physician's diagnosis, symptoms of reversible airflow obstruction, and current treatment.¹⁷ Enrollees 5 years or older also had a 12% or greater improvement in postbronchodilator FEV₁ from baseline or a methacholine PC₂₀ value of less than 16 mg/mL. Severe asthma was defined according to American Thoracic Society (ATS)¹⁸ and European Respiratory Society Task Force¹⁹ guidelines as either controlled or uncontrolled despite 6 months of treatment by an asthma specialty physician with high-dose inhaled or systemic corticosteroids and a second controller. For enrollees less than 6 years of age, severe asthma was defined by treatment with high-dose corticosteroids according to the ATS/European Respiratory Society high-dose definitions for children.¹⁹ Lung function was measured according to ATS guidelines for precision and reproducibility and expressed according to standard population reference values.²⁰ Criteria for poor asthma control included Asthma Control Test or the Childhood Asthma Control Test scores of less than 20 units.²¹ Exclusion criteria included a diagnosis of significant congenital or medical disorders apart from asthma.

³He MRI of the chest

³He MRI studies were performed within a few weeks of enrollment according to a protocol developed for children. Inhaled β -agonists were withheld for 4 hours before the MRI procedure. Enrollees were evaluated on the day of imaging for clinical stability by a research nurse coordinator. Criteria for exclusion were exposure to oral corticosteroids in the past 4 weeks before imaging, conditions for which an MRI procedure was contraindicated, the presence of any non-MRI-compatible metallic materials in the body, chest circumference larger than the MRI coil, and self-reported pregnancy. Children underwent the imaging protocol without sedation by using a 1.5-T commercial whole-body scanner (TIM Avanto; Siemens Medical Solutions, Malvern, Pa) and either a flexible, vest-shaped chest radio frequency (RF) coil (Clinical MR Solutions, Brookfield, Wis) or a fixed geometry RF coil (Rapid Biomedical, Rimpar, Germany) tuned to the ³He resonant frequency. Details of the ³He MRI image acquisition procedures are found in the [Methods](#) section in this article's Online Repository at www.jacionline.org.

Quantification of lung volume compartments

Lung images were subdivided into regions defined by the level of ³He ventilation by using an automated scoring platform.^{22,23} Six investigators individually traced whole-lung masks for each image, which were then combined into a single mask to help minimize rater bias,²⁴ to define the outline of the lungs. Bias correction was next performed to remove artifacts in the images caused by imperfections in the RF coil homogeneity and prepare the images for automated scoring.¹⁶ The lung images then underwent automated computer scoring in which the relative signal intensity in each voxel was used to classify the voxel into one of 4 classes: ventilation defect (black areas), hypoventilated (gray areas), ventilated (white areas), and well ventilated (bright white areas).²⁵ The sum of all of the voxels in each class times the voxel volume is the volume of the lung in each class. The sum of the 4 class volumes equaled the total lung volume for any one participant, and each of the volumes was expressed as a ratio of its value to total lung volume. The coinvestigator performing the image analysis (N.J.T.) was not aware of the clinical features and asthma severity of the participants.

Data analysis

Ventilation volumes and clinical variables were analyzed for normal distribution by using frequency histogram plots. Results were stratified for comparison based on tercile distribution (lower, middle, and upper) of the ventilation defect volume to total lung volume ratio. Differences in nonnormally distributed measurements across the terciles were tested by using the Kruskal-Wallis test and for normally distributed measurements by using ANOVA with adjustment for more than 2 groups with the *post hoc* Bonferroni adjustment. Nonscaled variables were tested with contingency tables and the Pearson χ^2 statistic. The Spearman rho statistic was used to test correlations among nonnormally distributed data.

RESULTS

Thirty-one children (median age, 10 years; minimum age, 3 years; maximum age, 17 years) were enrolled. The features of enrollees ([Table 1](#)) were similar to those of children who were not enrolled (results not shown). The sample included 12 (39%) of 31 preschool children (3-6 years of age) and 19 (61%) of 31 school-age children (7-17 years of age). The free-breathing protocol allowed safe acquisition (ie, no adverse events) of high-quality images, even in preschool children with severe asthma ([Fig 1](#)). Ventilation defects were classified according to 3 general patterns: no visible ventilation defect (7/31 [23%]), diffuse small defect (9/31 [29%]), and large focal defect (15/31 [48%], see [Fig E1](#) in this article's Online Repository at www.jacionline.org). Large ventilation defects were found only in children with severe asthma (n = 15/20 [75%]), and none (0/11) were found in patients with mild-to-moderate asthma ($P < .001$).

Ventilation volume compartments

Automated analysis of the corrected grayscale MRI defined 4 ventilation volumes labeled as red/green/blue/yellow regions in the image sections ([Fig 2](#)). A composite image derived from the individual hand-scored images defined a "ground-truth" image for each subject. The automated image corresponded to this ground-truth image better than any individual hand-scored image. The 4 volumes were expressed according to the percentage of total lung volume and adjusted for height (see [Table E1](#) in this article's Online Repository at www.jacionline.org). The ventilation defect volume percentage distributed asymmetrically to lower values (see [Fig E2](#) in this article's Online Repository at www.jacionline.org).

TABLE I. Features of the study sample stratified by the ventilation defect/total lung volume tercile distribution

Variable	Sample	Lower tercile (0.13% to 0.81%)	Middle tercile (1.0% to 2.1%)	Upper tercile (2.5% to 11.6%)	P value
No.	31	10	11	10	
Enrollment age (y)	10 (3-17)	9 (4-16)	11 (3-17)	9 (3-17)	.97 [†]
Male sex, no. (%)	21 (68)	7 (70)	7 (64)	7 (70)	.94
African American, no. (%)	10 (32)	2 (20)	4 (36)	4 (40)	.48
Body mass index (kg/m ²)*	18.3 (14.7-33.6)	18.5 (17.1-29.0)	17.5 (14.7-33.6)	19.6 (14.7-31.5)	.77
Birth gestational age (mo)	39 (24-39)	38 (28-39)	38 (27-39)	39 (24-39)	.66
Age at first wheeze (mo)	12 (2-108)	12 (2-60)	12 (2-24)	8 (2-108)	.89
Hospital admissions for asthma (no.)	1 (0-10)	1 (0-6)	1 (0-10)	6 (1-6)	.16
Any ICU admission for asthma, no. (%)	8 (26)	2 (20)	2 (18)	4 (40)	.46
Any past pneumonia, no. (%)	26 (84)	8 (80)	10 (91)	8 (80)	.73
Family history of asthma, no. (%)	24 (77)	9 (90)	8 (73)	7 (70)	.51
Daily ICS dose (μg fluticasone equivalent)	800 (0-2000)	445 (0-920)	460 (320-2000)	860 (80-1600)	.08
High-dose ICS, no. (%)	25 (81)	6 (60)	10 (91)	9 (90)	.13
Daily prednisone, no. (%)	7 (23)	0 (0)	2 (18)	5 (50)	.03
Daily LABA, no. (%)	22 (71)	5 (50)	8 (73)	9 (90)	.14
Montelukast, no. (%)	20 (64)	5 (50)	8 (73)	7 (70)	.50
No. of daily controllers	3 (1-4)	2 (1-4)	3 (1-4)	4 (1-4)	.05
cACT/ACT score	12 (8-25)	14 (11-25)	11 (8-23)	12 (9-14)	.21
Asthma event score	0.83 (0.0-3.0)	0.83 (0.25-1.5)	0.66 (0.0-2.8)	0.87 (0.0-3.0)	.84
Severe asthma, no. (%)	20 (64)	3 (30)	7 (64)	10 (100)	.005

cACT/ACT, Childhood Asthma Control Test/Asthma Control Test; ICS, inhaled corticosteroid; ICU, intensive care unit.

*Scaled variables shown as medians (minimum-maximum values).

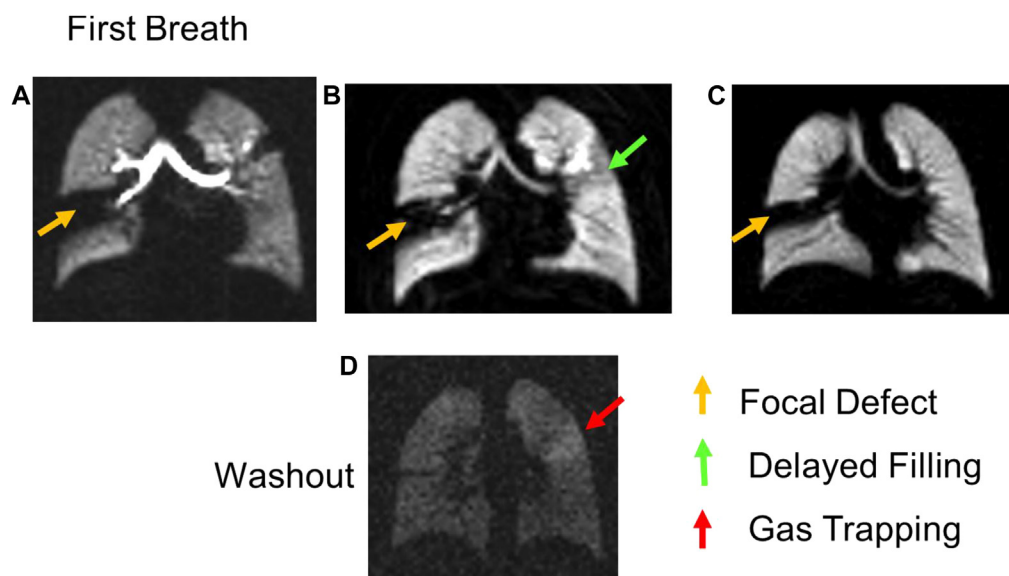
[†]Kruskal-Wallis test corrected for multiple group comparisons.

FIG 1. ³He lung MRI in a 4-year-old with severe asthma. This *grayscale* image illustrates the spectrum of signal intensities attained in a small child with the free-breathing acquisition protocol. **A**, Note that with the first inhalation of ³He, a large focal ventilation defect is visible in the *right middle* and *right lower* lobes. **B**, With subsequent inhalations, the defect persists, and there is delayed filling of a region in the *left upper* lobe. **C** and **D**, The region with delayed filling eventually fills (Fig 1, **C**) and during the washout phase (Fig 1, **D**) demonstrates relatively “bright” signal intensity consistent with trapped ³He gas.

Characteristics of the study participants were stratified according to the tercile distribution of the defect volume percentage to identify the clinical correlates of ventilation defects (Table I). Children with ventilation defect volume percentages in the upper tercile had more severe asthma, received more daily prednisone, and took more daily controller medications compared with

children with ventilation defect volume percentages in the middle or lower terciles. Children with ventilation defect volume percentages in the upper and middle terciles had lower FEV₁ percent predicted, FEV₁/FVC percent predicted, and forced expiratory flow rate from 25% to 75% of expired volume (FEF₂₅₋₇₅) percent predicted values than participants in the lower tercile (Table II).

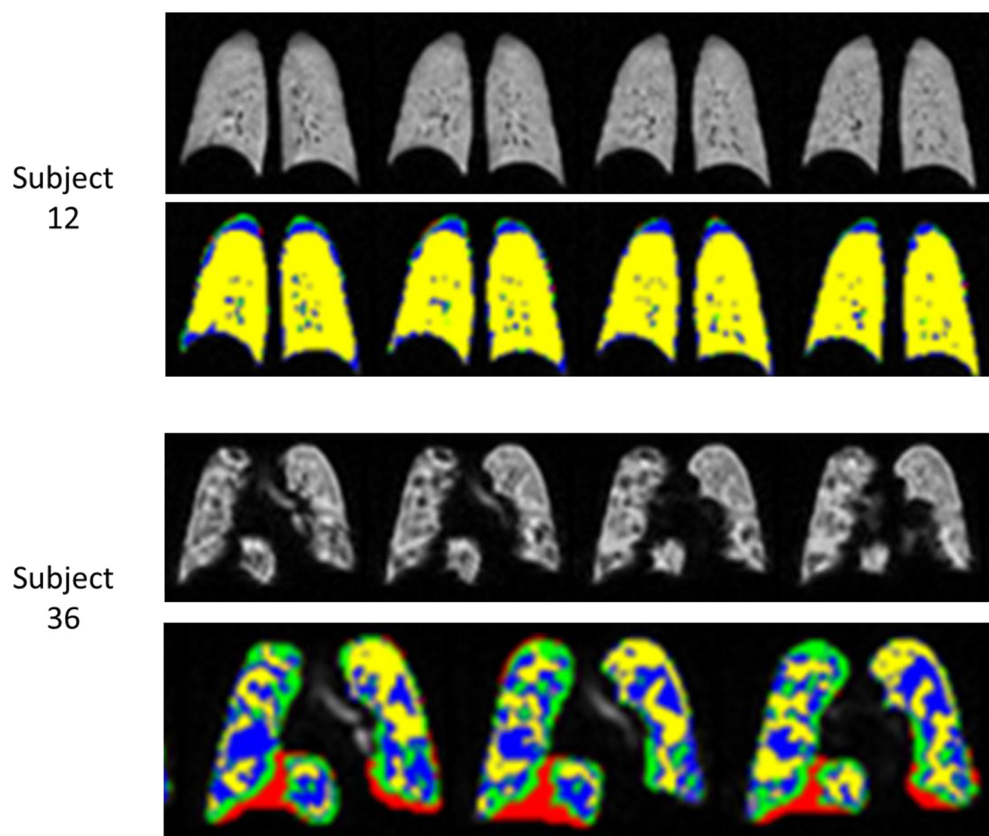


FIG 2. Coronal ^3He lung MRI slices in 2 asthmatic children. The *grayscale* images are shown above the corresponding labeled images, which have undergone automated analysis of the ^3He signal intensity. The ventilation defect volume is labeled *black* on the *grayscale* images and *red* on the automated images, the hypoventilated volume is *gray* on the *grayscale* images and *green* on the automated images, the ventilated volume is *white* on the *grayscale* images and *blue* on the automated images, and the well-ventilated volume is *bright white* on the *grayscale* images and *yellow* on the automated images. Subject 12 has mild-to-moderate asthma and normal lung function. The *grayscale* images show *primarily white* to *bright white* regions, and the automated images show mostly *yellow* regions. This subject had a relatively low defect volume to total lung volume ratio of 0.3%. By contrast, subject 36 has severe asthma, with significant airflow obstruction at baseline. The MRI shows visible contrasts in both the *grayscale* and automated images. Note the relative abundance of *red* and *green* regions on the automated images compared with subject 12. This subject had a relatively high defect volume to total lung volume ratio of 8.6%.

To further evaluate the effects of airflow limitation on distribution of ventilation, we compared lung volume compartments according to the tercile distribution of the FEV_1/FVC ratio. Children with FEV_1/FVC percentages in the lower tercile (<66% of predicted value) had significantly higher ventilation defects and hypoventilated and ventilated volume ratios but lower well-ventilated to total lung volume ratios versus children with FEV_1/FVC percent predicted values in the middle and upper terciles ($P < .05$ for all comparisons, Fig 3).

For markers of atopic inflammation, the number of positive allergens and peripheral blood eosinophil percentages were significantly higher in children whose ventilation defect volume percentages distributed to the upper tercile (Table III). However, the ventilation defect volume percentage was no greater in children sensitized to 1 or more allergens versus those with no allergen sensitization and was no greater in children with sensitization to 1 or more foods versus those with no food sensitization (see Table E2 in this article's Online Repository at www.jacionline.org).

Correlations between ventilation defects and clinical features of asthma

There were significant correlations (Fig 4 and Table IV) between the ventilation defect volume percentage and daily inhaled corticosteroid doses, numbers of asthma controllers, and Asthma Control Test/Childhood Asthma Control Test scores. Lung function variables with significant correlations with the ventilation defect percentages included FEV_1 , FEV_1/FVC , and FEF_{25-75} percentages but not FVC percentages. Markers of inflammation that correlated significantly with ventilation defect percentages included peripheral blood eosinophil percentages and total numbers of peripheral eosinophils.

Clinical features and ventilation volume ratios by asthma severity

Children with severe asthma ($n = 20$) had significantly more hospitalizations and more intensive care admissions and received more daily controller therapies and more maintenance prednisone

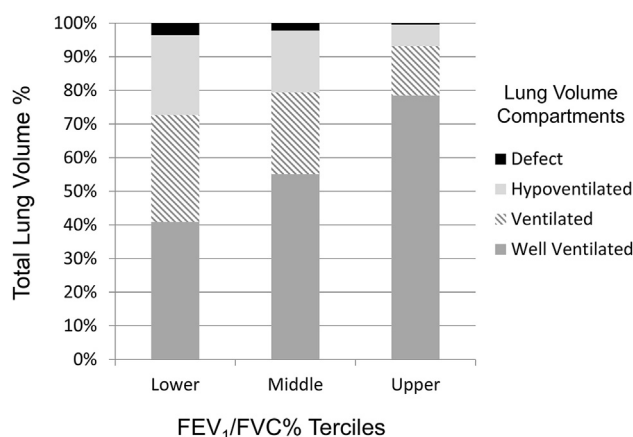
TABLE II. Lung function stratified by the ventilation defect/total lung volume tercile distribution

Variable	Sample	Lower tercile	Middle tercile	Upper tercile	P value
No.	31	10	11	10	
Sao ₂ in room air (%)*	98 ± 1	97 ± 1	98 ± 2	98 ± 32	.83
No.	16	6	6	4	
FEV ₁ (L)	1.87 ± 0.68	2.32 ± 0.64	1.78 ± 0.61	1.33 ± 0.49	.07
FEV ₁ (% predicted)	79 ± 24	102 ± 9	64 ± 6	65 ± 32	.004†
FVC (L)	2.75 ± 1.29	2.88 ± 1.04	3.08 ± 1.78	2.08 ± 0.61	.49
FVC (% predicted)	99 ± 25	109 ± 5	94 ± 24	91 ± 41	.47
FEV ₁ /FVC ratio	0.70 ± 0.13	0.82 ± 0.08	0.63 ± 0.12	0.63 ± 0.07	.008†
FEV ₁ /FVC ratio (% predicted)	78 ± 14	91 ± 9	70 ± 13	70 ± 9	.006†
FEF ₂₅₋₇₅ (L/s)	1.56 ± 0.94	2.49 ± 0.79	1.13 ± 0.29	0.84 ± 0.47	.004†
FEF ₂₅₋₇₅ (% predicted)	56 ± 38	93 ± 37	32 ± 5	32 ± 19	.007†

Sao₂, Arterial oxygen saturation.

*Mean ± 1 SD.

†P < .05, lower tercile versus middle and upper terciles, ANOVA corrected for multiple comparisons.

**FIG 3.** Bar plot of 4 ventilation volume compartments, as measured by means of automated analysis of the inhaled ³He MRI signal and stratified by the tercile distribution of the FEV₁/FVC ratio percent predicted in 31 asthmatic children. The volume compartments are expressed as the median values of the volume compartments expressed as percentages of the total lung volume. Children with FEV₁/FVC percent predicted in the lower tercile (<66% of predicted value) had significantly higher ventilation defect, hypoventilated, and ventilated volume ratios but lower well-ventilated to total lung volume ratios versus children with FEV₁/FVC percent predicted in the middle and upper terciles (P < .05 for all comparisons).

than children with mild-to-moderate asthma (n = 11, see Table E3 in this article's Online Repository at www.jacionline.org). Children with severe asthma had lower FEV₁, FEV₁/FVC, and FEF₂₅₋₇₅ percent predicted but no difference in FVC percent predicted versus children with mild-to-moderate asthma. There was no difference in total lung volume or lung volume adjusted by height in children with severe versus those with mild-to-moderate asthma. However, children with severe asthma had significantly higher ventilation defect, higher hypoventilated, higher ventilated, and lower well-ventilated to total lung volume ratios compared with volumes in children with mild-to-moderate asthma (see Fig E3 in this article's Online Repository at www.jacionline.org).

DISCUSSION

We report that the magnitude of the lung ventilation defect percentages in children correlates significantly with asthma severity, symptom control scores, treatment, lung function, and

biomarkers of atopy. Furthermore, this is the first report of lung volume compartments measured by means of automated analysis of the inhaled ³He signal in asthmatic children. We learned that the relative contribution of the ventilation defect and hypoventilated volume to total lung volume, both of which are estimates of the degree of ventilation heterogeneity, were greatest in children with severe asthma, and both volumes increased in children with airflow obstruction, as indicated by low FEV₁/FVC ratios. Thus precise measurement of ventilation volumes by hyperpolarized noble gas MRI not only resolved the spatial and temporal characteristics of gas distribution in asthmatic children but also was informative in regard to asthma severity and its clinical features.

Hyperpolarized gas imaging has significant promise as a safe and innovative clinical tool to noninvasively assess the regional distribution of ventilation in adults and children with asthma. This is possible through recent advances in automated signal analysis, as described in this and other reports,¹⁶ wherein the intensity of the inhaled ³He signal in airspaces is quantified to precisely map regional volume compartments and oxygen tension.²⁶ In the past, simple computer-assisted systems^{27,28} or hand counts of visual defects were used to estimate the ventilation defect volume.² In this study we compared hand scoring of visible defect regions using manual segmentation to an automated analysis platform and found that the automated analysis yielded volume estimates that corresponded best to the ground-truth or gold standard composite image. These automated scoring methods are important insofar as they could be used to facilitate rapid conversion of complex hyperpolarized gas signal data into volume compartments for clinical applications.

We found differences in the patterns and clinical correlates of ventilation defects in asthmatic children compared with those reported previously in adults. Although the fraction of asthmatic children with visible defects was 77% versus 65% in adults, the ventilation defect percentage was comparable in the 2 samples: 2.3% in children versus a range of 1.3% to 4.3% in adults.¹² Svenningsen et al¹² found that adults with visible defects were significantly older than those with no defects. However, we found no significant correlation between age and ventilation defect percentage in asthmatic children. In our study asthma severity highly informed the prevalence of defects in children, but severe asthma was more prevalent in school-age versus preschool enrollees. Our study was not powered to determine the effects of age on ventilation defect percentages in children; hence this

TABLE III. Markers of atopic inflammation stratified by ventilation defect/total lung volume tercile distribution

Variable	Sample	Lower tercile	Middle tercile	Upper tercile	P value
No.	27	8	9	10	
Serum IgE* (IU/mL)	210 (1-3638)	266 (24-3638)	113 (1-594)	637 (21-2047)	.07
No. of positive* allergens	2 (0-16)	1 (0-8)	1 (0-8)	8 (0-16)	.016†
Peripheral blood eosinophil count* (%)	4 (0-18)	1 (0-6)	6 (2-14)	8 (0-18)	.03‡
Total blood eosinophil* count (cells/ μ L)	0.42 (0.0-2.05)	0.21 (0.0-0.47)	0.74 (0.17-2.05)	0.61 (0.0-1.82)	.08
Self-reported wheeze after allergen (%)	21 (70)	6 (60)	6 (60)	9 (90)	.240
Atopic, no. (%)	22 (71)	7 (70)	6 (55)	9 (90)	.20

*Median (minimum-maximum) values.

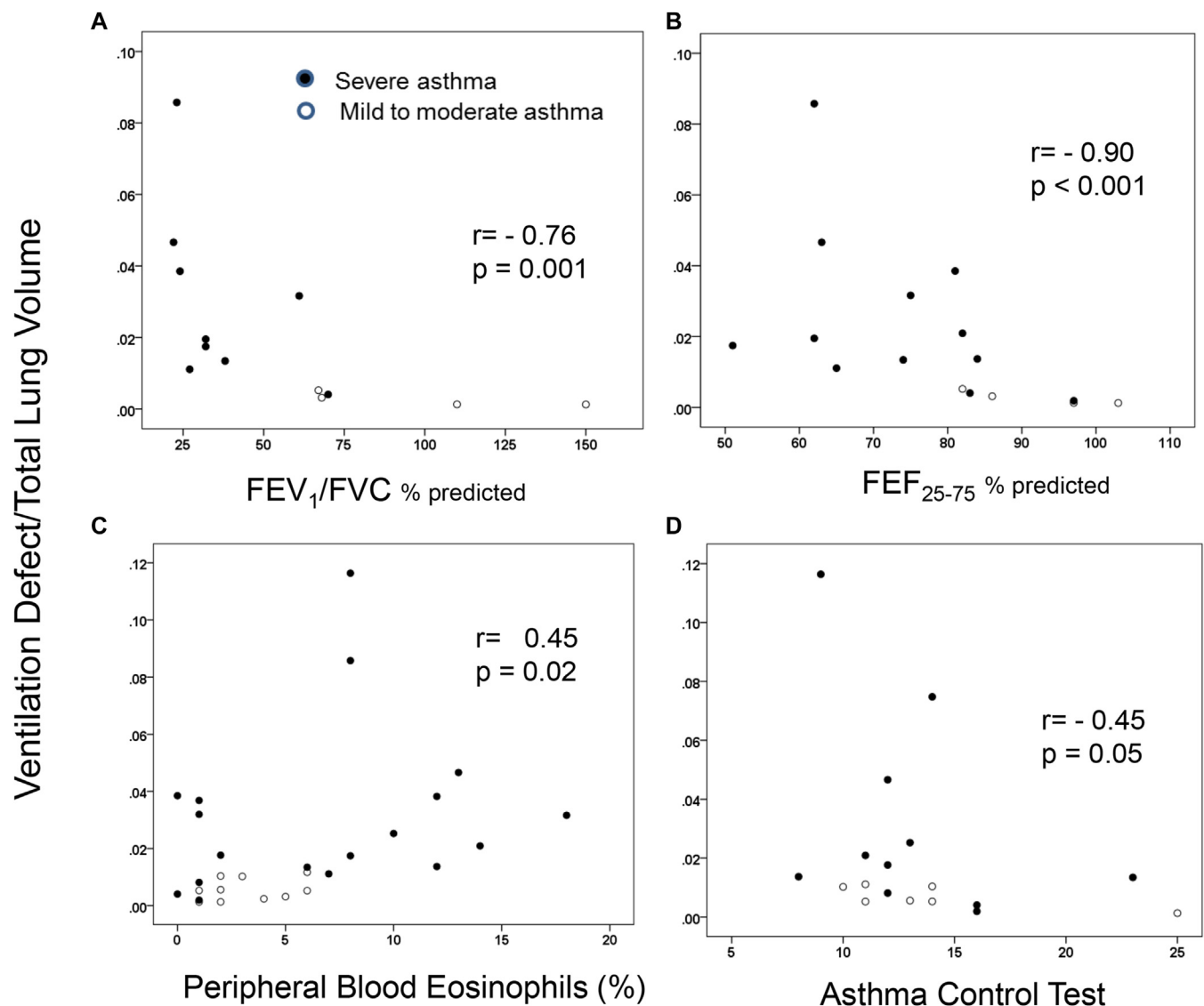
† $P < .05$, lower tercile versus middle and upper terciles, Kruskal-Wallis test corrected for multiple comparisons.‡ $P < .05$, lower tercile versus upper tercile, Kruskal-Wallis test corrected for multiple comparisons.**FIG 4.** Scatter plots showing correlation between the ventilation defect volume ratio with selected variables in asthmatic children. **A** and **B**, There was a strong inverse correlation between ventilation defect volume ratio and FEV₁/FVC percent predicted (Fig 4, **A**) and FEF₂₅₋₇₅ percent predicted (Fig 4, **B**). **C** and **D**, Ventilation defect volume ratio also correlated significantly with other clinical markers, including blood eosinophil percentages (Fig 4, **C**) and Asthma Control Test scores (Fig 4, **D**).

TABLE IV. Correlation of ventilation defect/total lung volume ratio with clinical features of asthma in children

Variable	Spearman rho	P value
Age	0.09	.61
Body mass index	−0.01	.99
Birth gestational age	0.19	.31
Age at first wheeze	−0.02	.91
Daily ICS dose	0.51	.003
No. of daily controllers	0.52	.003
Daily prednisone dose	0.54	.11
cACT/ACT scores	−0.45	.05
Asthma event score	0.09	.63
Sao ₂	−0.18	.35
FEV ₁	−0.71	.002
FVC	−0.47	.08
FEV ₁ /FVC	−0.76	.001
FEF ₂₅₋₇₅	−0.90	.0001
Serum IgE	0.23	.24
No. of positive allergens	0.33	.10
Peripheral eosinophils (%)	0.45	.02
Total blood eosinophils	0.39	.04

cACT/ACT, Childhood Asthma Control Test/Asthma Control Test; ICS, inhaled corticosteroid; Sao₂, arterial oxygen saturation.

question is not resolved. de Lange et al¹⁰ reported a significant negative correlation between the numbers of lung defects per image slice and FEV₁/FVC and FEF₂₅₋₇₅ values in adults. Svenningsen et al¹² found no difference in FEV₁ percentages in asthmatic patients with and without apparent ventilation defects but reported a strong negative correlation with ventilation defects and FEV₁ percentages. In contrast, we found that the relationship between defect percentages and asthma features was strongest for the FEF₂₅₋₇₅ percent predicted. This result and those of Svenningsen et al¹² and de Lange et al¹⁰ support the evolving evidence pointing to pathologic changes in the lung periphery as contributing in an important way to the maldistribution of ventilation and the pathobiology of asthma.²⁹

The present study has design and sample size disadvantages that limit broad application of the findings to asthmatic patients. Foremost, the study sample features are enriched in children with severe and poorly controlled asthma. The burden of inhaled corticosteroid exposure was high and asthma control was poor, even in the mild-to-moderate group, although enrollees with mild-to-moderate asthma had relatively normal lung function compared with participants with severe asthma. As a result, we might overestimate the magnitude of ventilation defect percentages in the general population of asthmatic children. This is the likely reason our findings are different from those of Cadman et al,¹¹ who studied older children with mild-to-moderate asthma and less airflow limitation based on a relatively high FEV₁/FVC ratio in their sample.

A second limitation of the study is that there is no control group for quantification of ventilation defects in healthy children. Apparent ventilation defects were not identified in healthy adults in the report by Svenningsen et al,¹² but in reports from de Lange et al,¹⁰ small peripheral ventilation defects were commonly seen in healthy adult volunteers. The prevalence and volume of ventilation defects identified by means of ³He MRI in healthy children have not been widely reported.

A third limitation of the study is that it is cross-sectional, and thus it is not possible to infer causality between the defects and

clinical features or *vice versa*. This will require an interventional study designed to reverse ventilation defects to see which, if any, clinical features of asthma improve. The fourth disadvantage is that many of the images were obtained in younger children who were tidally breathing and unable to perform a standard volume maneuver. For this reason, we did not attempt to measure the helium diffusion path constant, as has been reported in older children with asthma by Cadman et al.¹¹

The feasibility of ³He MRI as a tool in the clinical management of asthmatic patients has promise but will require further clinical studies. This will depend in part on advances in automated signal analysis, as we have reported, so that quantification of lung volume compartments will be accessible to practitioners. MRI has significant advantages over chest CT insofar as it does not involve exposure to ionizing radiation, a highly important consideration in children. Hyperpolarized noble gas lung MRI might have particular application in evaluation of the peripheral airways in asthmatic patients.³⁰ Spirometry, forced oscillometry, and nitrogen clearance are available methods to assess the peripheral lung, but these methods address global lung function and are less sensitive to regional patterns of disease. The advent of hyperpolarized gas methods has changed the way we think about lung function in asthmatic patients, from an understanding of asthma as a uniform disorder of airways obstruction to a disorder more akin to regional ileitis with patchy involvement. Inhaled corticosteroids, especially those with coarse particle features, likely do not penetrate ventilation defect regions in asthmatic patients.³¹ Inhaled ³He lung MRI is ideally suited to evaluate regional ventilation changes after treatment, as has been shown recently for bronchial thermoplasty.³² By means of assessment of the ³He diffusion path, lung MRI has also been used to assess lung development and alveolar growth in ex-premature children.³³

We acknowledge the efforts of Kristin Wavell and Donna Wolf, PhD, who participated in validation of the manual segmentation volume estimates with the Advanced Normalization Tools (ANTs)-derived volumes. Denise Thompson-Batt, RRT, assisted in the recruiting and enrollment of participants in the study.

REFERENCES

- de Lange EE, Mugler JP 3rd, Brookeman JR, Knight-Scott J, Truwitt JD, Teates CD, et al. Lung air spaces: MR imaging evaluation with hyperpolarized ³He gas. *Radiology* 1999;210:851-7.
- Altes T, Powers P, Knight-Scott J, Rakes G, Platts-Mills T, Lange E, et al. Hyperpolarized ³He MR lung ventilation imaging in asthmatics: preliminary findings. *J Magn Reson Imaging* 2001;13:378-84.
- Castro M, Fain SB, Hoffman EA, Gierada D, Erzurum SC, Wenzel S. Lung imaging in asthma: the picture is clearer. *J Allergy Clin Immunol* 2011; 128:467-78.
- Irvin CG, Bates JHT. Physiologic dysfunction of the asthmatic lung. What's going on down there anyway? *Proc Am Thorac Soc* 2009;6:306-11.
- Venegas JG, Winkler T, Musch G, Melo MF, Layfield D, Tgavalekos N, et al. Self-organized patchiness in asthma as a prelude to catastrophic shifts. *Nature* 2005; 434:777-82.
- Harris RS, Winkler T, Tgavalekos N, Musch G, Melo MCFV, Schroeder T, et al. Regional pulmonary perfusion, inflation, and ventilation defects in bronchoconstricted patients with asthma. *Am J Respir Crit Care Med* 2006;174:245-53.
- Fain SB, Gonzalez-Fernandez G, Peterson ET, Evans MD, Sorkness RL, Jarjour NN, et al. Evaluation of structure-function relationships in asthma using multi-detector CT (MDCT) and hyperpolarized (HP) ³He MRI. *Acad Radiol* 2008;15:753-62.
- de Lange EE, Altes TA, Patrie JT, Battiston JJ, Juersivich AP, Mugler JP 3rd, et al. Changes in regional airflow obstruction over time in the lungs of patients with asthma: evaluation with ³He MR imaging. *Radiology* 2009;250:567-75.

9. Costella S, Kirby M, Maksym GN, McCormack DG, Paterson NA, Parraga G. Regional pulmonary response to a methacholine challenge using hyperpolarized ^3He magnetic resonance imaging. *Respirology* 2012;17:1237-46.
10. de Lange EE, Altes TA, Patrie JT, Gaare JD, Knake JJ, Mugler JP, et al. Evaluation of asthma with hyperpolarized ^3He MRI: correlation with clinical severity and spirometry. *Chest* 2006;130:1055-62.
11. Cadman RV, Lemanske RF, Evans MD, Jackson DJ, Gern JE, Sorkness RL, et al. Pulmonary ^3He magnetic resonance imaging of childhood asthma. *J Allergy Clin Immunol* 2013;131:369-76.
12. Svenningsen S, Kirby M, Starr D, Coxson HO, Paterson NA, McCormack DG, et al. What are ventilation defects in asthma? *Thorax* 2014;69:63-71.
13. Macleod KA, Horsley AR, Bell NJ, Greening AP, Innes JA, Cunningham S. Ventilation heterogeneity in children with well controlled asthma with normal spirometry indicates residual airways disease. *Thorax* 2009;64:33-7.
14. Papastamelos C, Panitch HB, England DE, Allen JL. Developmental changes in chest wall compliance in infancy and early childhood. *J Appl Physiol* 1995;78:179-84.
15. Altes TA, Mata J, de Lange EE, Brookeman JR, Mugler JP III. Assessment of lung development using hyperpolarized ^3He diffusion MR imaging. *Magn Reson Imaging* 2006;24:1277-83.
16. Tustison NJ, Altes TA, Song G, de Lange EE, Mugler JP, Gee JC. Feature analysis of hyperpolarized ^3He MRI: a study of asthmatics versus non-asthmatics. *Magn Reson Med* 2010;63:1448-55.
17. National Asthma Education and Prevention Program. Guidelines for the diagnosis and management of asthma. Expert panel report 3. Bethesda: National Heart, Lung, and Blood Institute, National Institute of Health; 1997.
18. American Thoracic Society. Proceedings of the American Thoracic Society Workshop on Refractory Asthma: current understanding, recommendations, and unanswered questions. *Am J Respir Crit Care Med* 2000;162:2341-51.
19. Chung KF, Wenzel SE, Brozek JL, Bush A, Castro M, Sterk P, et al. International ERS/ATS consensus definition, mechanisms, evaluation and treatment of severe asthma. *Eur Respir J* 2014;43:343-73.
20. Hankinson JL, Odencrantz JR, Fedan KB. Spirometric reference values from a sample of the general U.S. population. *Am J Respir Crit Care Med* 1999;159:179-87.
21. National Institutes of Allergy, Asthma, and Infectious Diseases. Standardizing asthma outcomes in clinical research: report of the asthma outcomes workshop. *J Allergy Clin Immunol* 2012;129(suppl):S34-8.
22. Tustison NJ, Avants BB, Flors L, Altes TA, de Lange EE, Mugler JP, et al. Ventilation-based segmentation of the lungs using hyperpolarized ^3He MRI. *J Magn Reson Imaging* 2011;34:831-41.
23. Yushkevich PA, Piven J, Hazlett HC, Smith RG, Ho S, Gee JC, et al. User-guided 3D active contour segmentation of anatomical structures: significantly improved efficiency and reliability. *Neuroimage* 2006;31:1116-28.
24. Warfield SK, Zou KH, Wells WM. Simultaneous truth and performance level estimation (STAPLE): An algorithm for the validation of manual segmentation. *IEEE Trans Med Imaging* 2004;23:903-21.
25. Avants BB, Tustison N, Wu J, Cook PA, Gee JC. An open source multivariate framework of n-tissue segmentation with evaluation on public data. *Neuroinformatics* 2011;9:381-400.
26. Miller GW, Mugler JP 3rd, Altes TA, Cai J, Mata JF, de Lange EE, et al. A short-breath-hold technique for lung pO₂ mapping with ^3He MRI. *Magn Reson Med* 2010;63:127-36.
27. Parraga G, Ouriadvod A, Evans A, McKay S, Lam WW, Fenster A, et al. Hyperpolarized ^3He ventilation defects and apparent diffusion coefficients in chronic obstructive pulmonary disease: preliminary results at 3.0 tesla. *Invest Radiol* 2007;42:384-91.
28. Woodhouse N, Wild JM, Paley MNJ, Fichelle S, Said Z, Swift AJ, et al. Combined ^3He /proton magnetic resonance imaging measurement of ventilated lung volumes in smokers compared to never-smokers. *J Magn Reson Imaging* 2005;21:365-9.
29. Thompson BR, Douglass JA, Ellis MJ, Kelly VJ, O'Hehir RE, King GG, et al. Peripheral lung function in patients with stable and unstable asthma. *J Allergy Clin Immunol* 2013;131:1322-8.
30. Castro M, Woods J. Insights into pediatric asthma with hyperpolarized magnetic resonance imaging of the lung. *J Allergy Clin Immunol* 2013;131:377-8.
31. Goldin JG, Tashkin DP, Kleerup EC, Greaser LE, Haywood UM, Sayre JW, et al. Comparative effects of hydrofluoroalkane and chlorofluorocarbon beclomethasone dipropionate inhalation on small airways: assessment with functional helical thin-section computed tomography. *J Allergy Clin Immunol* 1999;104:S258-67.
32. Thomen RP, Sheshadri A, Quirk JD, Ellison HD, Szczesniak RD, Castro M, et al. Regional ventilation changes in severe asthma after bronchial thermoplasty. *Radiology* 2015;274:250-9.
33. Narayanan M, Beardsmore CS, Owers-Bradley J, Dogaru CM, Mada M, Ball I, et al. Catch up alveolarization in ex-preterm children: evidence from ^3He magnetic resonance. *Am J Respir Crit Care Med* 2013;187:1104-9.

METHODS

Description of clinical characterization procedures

Participants underwent a physical examination, detailed medical and treatment history, spirometry before and after bronchodilator, and studies for atopy, including total serum IgE measurement, measurement of specific IgE to a panel of inhalant and food allergens (Phadia ImmunoCAP; Pharmacia, Uppsala, Sweden), and a peripheral blood eosinophil count. The allergens tested included tree mix, grass mix, ragweed, molds, dust mite, cats, dogs, cock roach, peanut, egg white, soy, milk, and wheat. Participants withheld bronchodilator medications and had repeat spirometry on the day of the MRI. Long-term control was measured by using a composite index of episodes of poor asthma control termed the asthma event score. Episodes were scored as (1) mild exacerbations treated only with albuterol (score = 1/event), (2) exacerbations requiring prednisone bursts (score = 2/event), (3) emergency department visits (score = 3/event), hospital admissions (score = 4/event), and intensive care unit admissions (score = 6/event) 6 months before and 6 months after the MRI.

Details of ^3He MRI image acquisition procedures and breathing protocols

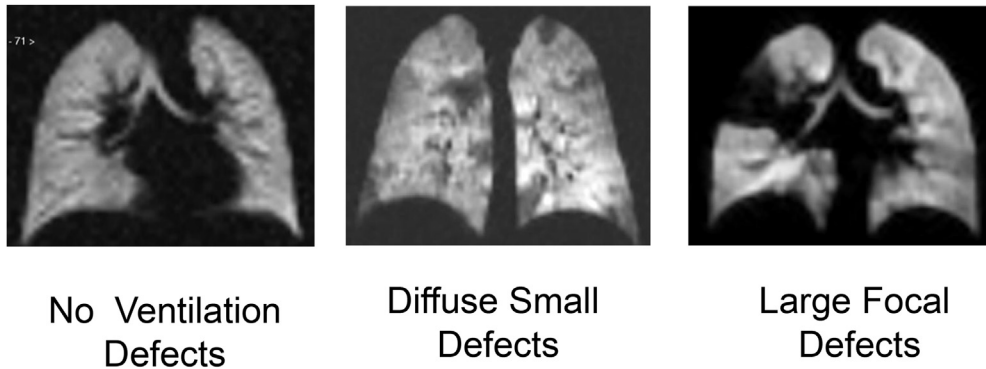
Helium gas was polarized with a home-built ^3He polarizer that used hybrid rubidium-potassium spin-exchange optical pumping with line-narrowed lasers.

Throughout the MRI session, heart rate and oxygen saturation level were monitored (model 3150 MRI Patient Monitor; Invivo Research, Gainesville, Fla). As part of the evaluation before the MRI scan, all subjects were asked to hold their breath for 10 seconds. Depending on whether they were able to

hold their breath, either a free-breathing acquisition or a breath-hold acquisition was performed. The free-breathing acquisition was optimized for speed to, in essence, freeze motion in children too young to perform breath-hold maneuvers. The breath-hold acquisition provided greater spatial resolution.

Free-breathing acquisition. The desired volume of ^3He (typically 0.5 L) was dispensed into a Tedlar bag (Jensen Inert Products, Coral Springs, Fla). The child was positioned supine on the scanner table with the ^3He RF coil around the chest. Contiguous, coronal, 2-dimensional, interleaved spiral ^3He images were acquired by using the following parameters: TR (repetition time)/TE (echo time), 8.1/0.9 ms (3 subjects) or 8.7/0.9 ms (2 subjects); flip angle, 20° ; voxel volume, $3.3 \times 3.3 \times 10 \text{ mm}^3$ or $2.0 \times 2.0 \times 10.0 \text{ mm}^3$; spiral interleaves, 13 or 20, plus 2 for a field map; and acquisition time, 0.12 seconds per slice or 0.19 seconds per slice. A field of view of at least 40 cm was used to ensure the child would be within the field of view, even if the child moved during acquisition. The pulse sequence was started just before gas administration, and the full set of coronal slices was acquired repeatedly (≥ 5 sets) because it could not be predicted in advance when the child would inhale a volume of ^3He sufficient for imaging. Imaging typically continued for several seconds during free breathing.

Breath-hold acquisition. A 1- or 2-L plastic bag (Jensen Inert Products) was filled with between 200 and 600 mL of ^3He and enough nitrogen to total approximately one third of the subject's FVC. Imaging was performed during an approximately 10-second breath hold immediately after the subject inhaled the gas mixture. Three-dimensional ^3He image sets of the lung were acquired by using a steady-state free precession (TrueFISP) with the following parameters: TR/TE, 1.9/0.8 ms (helium-3) or 1.8/0.7 ms (proton); flip angle, 9° ; and isotropic spatial resolution, 3.9 mm. Elliptical k-space sampling and the partial Fourier technique were used. A single set of whole-lung coronal images were acquired after gas inhalation.



Ventilation Defect Pattern	Total n = 31	Severe Asthma n = 20	Mild to Moderate Asthma n = 11
None n (%)	7 (23%)	0	7 (64%)
Diffuse small n (%)	9 (29%)	5 (25%)	4 (36%)
Large focal n (%)	15 (48%)	15 (75%)	0

FIG E1. Lung ventilation defect patterns and contingency tables of defect patterns by asthma severity identified by means of inhaled ^3He gas lung MRI. Large focal defects were found exclusively in children with severe asthma (n = 15/20) and not identified in children with mild-to-moderate asthma (n = 0/11; $P < .001$, Pearson χ^2 test).

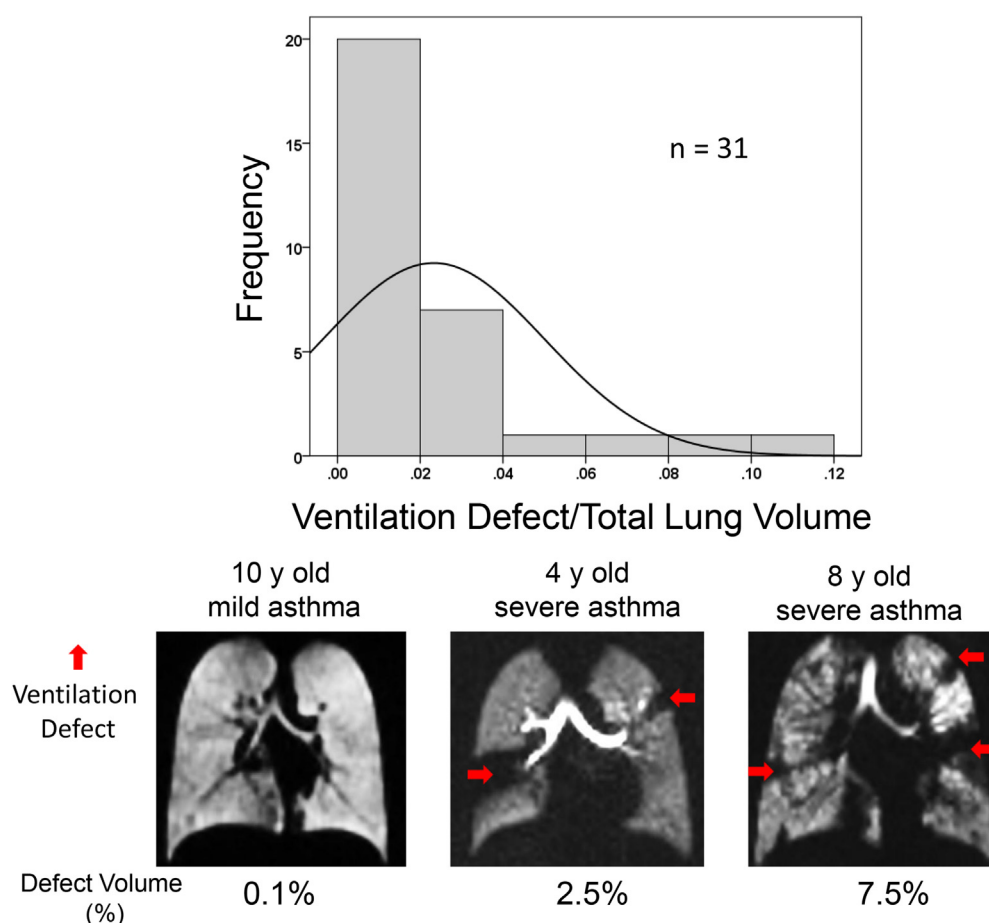


FIG E2. Frequency distribution plot of the ventilation defect/total lung volume in 31 asthmatic children. The x-axis scale is the absolute value of the defect volume to total lung volume fraction. Representative grayscale images from 3 asthmatic children are shown below the distribution plot. The corresponding ventilation defect volume ratio is expressed as the percentage of total volume for each child. *Red arrows* label ventilation defects.

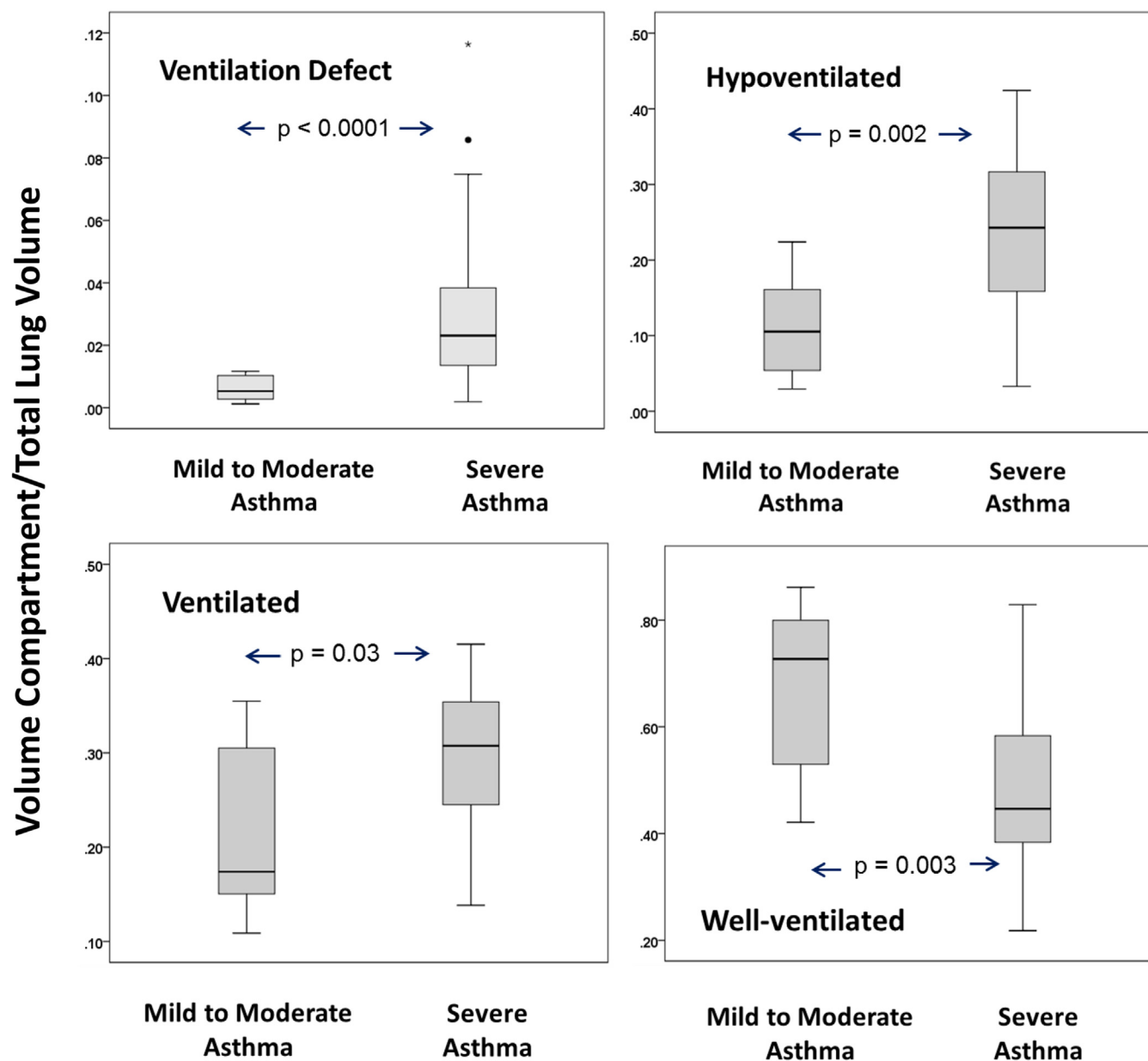


FIG E3. Box plots with CIs of lung volume compartments in children with severe ($n = 20$) and mild-to-moderate ($n = 11$) asthma defined by analysis of the inhaled ^3He signal. P values using the Mann-Whitney U test for 2 unpaired samples are shown.

TABLE E1. Lung volume compartments in 31 asthmatic children

Volume compartments	Mean $\pm$ SD	Median	Minimum value	Maximum value
Ventilation defect (L)	0.0477 $\pm$ 0.0544	0.02624	0.0027	0.2363
Ventilation defect/total lung (%)	2.3 $\pm$ 2.6	1.3	0.13	11.6
Hypoventilated (L)	0.3562 $\pm$ 0.2831	0.2868	0.0697	1.0997
Hypoventilated/total lung (%)	16.7 $\pm$ 9.0	16.1	2.8	34.9
Ventilated (L)	0.5479 $\pm$ 0.3439	0.3858	0.1800	1.5190
Ventilated/total lung (%)	26.6 $\pm$ 8.9	29.6	11.0	42.0
Well ventilated (L)	1.2810 $\pm$ 1.0654	0.9101	0.2105	4.7406
Well ventilated/total lung (%)	54.2 $\pm$ 18.6	50.5	21.8	86.1
Total lung	2.2329 $\pm$ 1.4460	2.0853	0.6270	7.4902
Total lung/height (L/m)	1.59 $\pm$ 0.76	1.54	0.600	4.150

TABLE E2. Allergen sensitization and lung ventilation compartments in asthmatic children

Volume compartment	No. of allergens	1-9 Allergens	10-16 Allergens	<i>P</i> value
No.	10	12	4	
Ventilation defect (L)	0.0128 (0.003-0.236)	0.0145 (0.003-0.097)	0.0771 (0.0317-0.109)	.18
Ventilation defect/total lung (%)	1.2 (0.2- 11.6)	1.0 (0.1-4.6)	3.5 (3.1-3.8)	.11

Results are median (minimum-maximum) values. *P* values were determined by using the Kruskal-Wallis test corrected for multiple group comparisons.

TABLE E3. Characteristics of children with severe (n = 20) and mild-to-moderate asthma (n = 11)

Variable	Severe asthma	Mild-to-moderate asthma	P value
Age (mo)	130 (37-210)	70 (40-198)	.09
Body mass index (kg/m ²)	18.9 (14.7-33.6)	17.2 (14.7-29)	.21
Birth gestational age (mo)	38 (24-42)	39 (36-39)	.48
Age at first wheeze (mo)	12 (2-108)	21 (2-60)	.23
No. of hospitalizations for asthma	2 (0-10)	0 (0-3)	.001
ICU admission, no. (%)	8 (40)	0	.015
Past pneumonia, no. (%)	17 (85)	9 (82)	.82
Daily ICS dose (fluticasone equivalent)	800 (80-2000)	440 (0-920)	.07
High-dose daily ICS, no. (%)	18 (90)	7 (64)	.07
No. of daily controllers (no./d)	3 (1-4)	2 (0-3)	.002
Daily prednisone, no. (%)	7 (37)	0	.021
Prednisone dose (mg)	17 (10-20)		.04
cACT/ACT scores	12 (8-23)	13 (10-25)	.93
Asthma event score	0.87 (0-3.0)	0.83 (0.25-2.8)	1.00
Sao ₂ (%)	98 (93-100)	98 (96-100)	.52
FEV ₁ (% predicted)	73 (33-132)	103 (90-111)	.01
FVC (% predicted)	93 (51-119)	102 (96-113)	.13
FEV ₁ /FVC (% predicted)	76 (62-103)	100 (80-109)	.02
FEF ₂₅₋₇₅ (% predicted)	46 (15-102)	100 (56-130)	.05
Serum IgE (IU/mL)	345 (21-3638)	146 (1-1126)	.14
No. of positive allergens	4 (0-16)	1 (0-8)	.36
Blood eosinophils (%)	7 (0-18)	2 (1-6)	.21
Total eosinophil count (cells/μL)	0.60 (0-2.0)	0.35 (0-0.78)	.28

All scaled variables are median (minimum-maximum) values of 2 group comparisons by using the Mann-Whitney *U* test.

cACT/ACT, Childhood Asthma Control Test/Asthma Control Test; ICS, inhaled corticosteroid; ICU, intensive care unit; Sao₂, arterial oxygen saturation.