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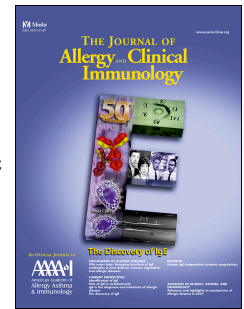
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# Quantitative CT imaging-based clustering differentiates asthmatic subgroups with distinctive clinical phenotypes



## Imaging characteristics

## Clinical characteristics

### Cluster 1

- Normal airway structure
- Increased lung deformation (Jacobian and ADI $\uparrow$ )

- Younger, early onset
- Nonsevere asthma
- Reversible lung function
- Easy to control asthma symptoms

### Cluster 2

- Airway luminal narrowing ( $D_h^*$  $\downarrow$ )
- No airway wall thickening (WT\*)
- Significant reduction of lung deformation (Jacobian and ADI $\downarrow$ )

- Nonsevere and severe asthma
- Persistently altered lung function
- Marginal to no inflammation
- Difficult to control asthma symptoms

### Cluster 3

- Airway wall thickening (WT\* $\uparrow$ )
- No airway luminal narrowing ( $D_h^*$ )
- Moderate reduction of lung deformation (Jacobian and ADI $\downarrow$ )

- Obese, female-dominant
- Severe asthma
- Reversible lung function
- Blood lymphopenia
- Difficult to control asthma symptoms

### Cluster 4

- Airway luminal narrowing ( $D_h^*$  $\downarrow$ )
- Significant reduction of lung deformation (Jacobian and ADI $\downarrow$ )
- Significant air-trapping (AirT% $\uparrow$ )

- Older, late onset, male-dominant
- Severe asthma
- Persistently altered lung function
- Neutrophilic-dominant inflammation
- Difficult to control asthma symptoms

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## ABSTRACT

**Background:** Imaging variables including airway diameter, wall thickness and air-trapping have been found to be important metrics when differentiating severe asthmatics from nonsevere asthmatics and healthy subjects.

**Objective:** The objective of this study was to identify imaging-based clusters and to explore the association of the clusters with existing clinical metrics.

**Methods:** We performed an imaging-based cluster analysis using quantitative computed tomography-based structural and functional variables extracted from the respective inspiration and expiration scans of 248 asthmatics. The imaging-based metrics included a broader-set of multiscale variables such as inspiratory airway dimension, expiratory air-trapping and registration-based lung deformation (inspiration vs. expiration). Asthma subgroups derived from a clustering method were associated with subject demography, questionnaire, medication history, and biomarker variables.

**Results:** Cluster 1 patients were early-onset younger nonsevere asthmatics with reversible airflow obstruction, who showed normal airway structure; Cluster 2 patients were a mix of nonsevere and severe asthmatics with marginal inflammation, who exhibited airway luminal narrowing without wall thickening. Cluster 3 and 4 patients were dominated by severe asthmatics. Cluster 3 patients were obese females with reversible airflow obstruction who exhibited airway wall thickening without airway narrowing. Cluster 4 patients were late-onset older males with persistent airflow obstruction, exhibiting significant air-trapping and reduced regional deformation. Clusters 3 and 4 patients also showed decreased lymphocyte and increased neutrophils, respectively.

**Conclusions:** Four image-based clusters were identified and shown to be correlated with clinical characteristics. Such clustering serves to differentiate asthma subgroups which may be used as a basis for the development of new therapies.

**Clinical Implications:**

The unique structural and functional alterations observed in each cluster provide new insights on the existing pathophysiology of asthma. The clustering membership employed in this analysis may prove to be useful for the development of effective interventions of asthmatics along with a future study.

**Capsule Summary:**

We identified four asthma subgroups using a cluster analysis composed of imaging variables, which were associated with clinical metrics. Identifying imaging-based clusters could enable practical cluster-based therapeutic interventions.

**Key words:** Computed tomography; image processing; severe asthma; air-trapping; image registration; luminal narrowing; wall thickening; airway circularity; cluster analysis; neutrophilic asthma

**Abbreviations used:**

SARP: Severe Asthma Research Program

FEV<sub>1</sub>: Forced expiratory volume in one second

QCT: Quantitative computed tomography

MICA: Multiscale imaging-based cluster analysis

LA: Airway luminal area

WA%: Airway wall area percentage, i.e., the ratio of wall area to total area

AirT%: Air-trapping percentage (residual gas at expiration)

NIH: National Institutes of Health

ATS: American Thoracic Society

PITT: The University of Pittsburgh

WSL: Washington University in St. Louis

WIS: The University of Wisconsin

FDR: False discovery rate

$\theta$ : Bifurcation angle between two daughter branches

$Cr$ : Airway luminal circularity

WT: Airway wall thickness

$D_h$ : Hydraulic luminal diameter

RMB: Right main bronchus

LMB: Left main bronchus

BronInt: Right intermediate bronchus

TriLLB: Trifurcation of left lower lobe

sRUL: Sub-grouped right upper lobe with branches of RB1 – 3 illustrated in Figure 1a

sRML: Sub-grouped right middle lobe with branches of RB4 – 5 illustrated in Figure 1a

sRLL: Sub-grouped right lower lobe with branches of RB6 – 10 illustrated in Figure 1a

sLUL: Sub-grouped left upper lobe with branches of LB1 – 5 illustrated in Figure 1a

sLLL: Sub-grouped left lower lobe with branches of LB6, LB8 – 10 illustrated in Figure 1a

- 94 TLC: Total lung capacity
- 95 FRC: Functional residual capacity
- 96 ADI: Anisotropic deformation index
- 97  $\Delta V_{\text{air}}^{\text{F}}$ : Lobar fraction of air volume change between TLC and FRC
- 98 Lung shape: Apical-basal distance over ventral-dorsal distance at TLC
- 99  $U/(M+L)/v$ : The ratio of air volume changes in upper lobes to those in middle and lower lobes
- 100 PCA: Principal component analysis
- 101 BMI: Body mass index
- 102 PFT: Pulmonary function test
- 103 AQLQ: Asthma quality of life questionnaire
- 104 ACQ: Asthma control questionnaire; ACQ-6: without  $FEV_1$ , ACQ-7: with  $FEV_1$
- 105 ACT: Asthma control test
- 106  $PC_{20}$ : Concentration of methacholine needed to produce a 20% fall in baseline  $FEV_1$
- 107 FeNO: Fraction of exhaled nitric oxide
- 108 BAL: bronchial alveolar lavage
- 109 OCS: Oral corticosteroids
- 110 ICS: Inhaled corticosteroids
- 111 LABA: Long-acting  $\beta_2$ -adrenergic agonists
- 112 SABA: short-acting  $\beta_2$ -adnergic agonists
- 113 LTRA: Leukotriene receptor antagonist
- 114 SPT: skin prick test
- 115 IgE: Immunoglobulin E
- 116 WBC: White blood cell
- 117

## INTRODUCTION

As a multicenter study of severe asthma, the Severe Asthma Research Program (SARP) has established genetic, environmental and clinical variables for characterization of asthma phenotypes, leading to a better understanding of asthma pathology (1-6). For instance, Moore et al. (2) have utilized only clinical variables in a cluster analysis to derive clinical clusters. Their study has also defined “simple” clinical clusters, 80% accuracy of original clusters, with only three factors including the onset age of asthma, and pre- and post-bronchodilator forced expiratory volume in one second (FEV<sub>1</sub>). Moore et al. (7) recently performed another cluster analysis using inflammatory biomarkers in addition to clinical characteristics, leading to 4 asthma sub-groups. While quantitative computed tomography (QCT) of lungs has been an important component of SARP, imaging variables have not been explored as the basis for identifying patient clusters. Only a limited set of available QCT metrics has been exploited with not only SARP but most of the multi-center studies incorporating QCT of the lungs. In this context, the objective of this study was to develop imaging-based asthma clusters using a variety of imaging-based variables reflecting airway and parenchymal pathologies at multiscale levels.

Multiscale imaging-based cluster analysis (MICA) uses a set of QCT-based segmental airway (local scale) as well as lung and lobar-based (global scale) density and shape-based variables (8-10) to identify clinically meaningful patient clusters. The rationale behind MICA is that structural and functional alterations may occur regionally and/or globally. Thus, to capture a wide spectrum of disease states, it is necessary to analyze variables at multiple scales. QCT (11-18) has identified structural and functional variables, which, in isolation, distinguish severe asthmatics from healthy and nonsevere asthmatic subjects, and further correlate with physiologic and clinical parameters. For instance, it has been found that luminal area (LA) (19) decreases, and airway wall area percentage (WA%) (20) and air-trapping percentage (AirT%) (21, 22) increase mostly in severe asthmatics, showing significant correlations with FEV<sub>1</sub>. The observed structural and functional alterations are aligned with the pathophysiology of asthma such as

airway narrowing, chronic inflammation, small airway disease and reduced elastic recoil (23, 24). A strength of imaging in the study of asthma is its ability to extract regional measurements because obstruction has been shown to be regionally heterogeneous (21, 25). In addition these non-invasively acquired imaging biomarkers can be followed for the longitudinal assessment of therapies directed against bronchial wall thickening and air trapping.

However, the use of existing imaging metrics across multi-center studies have been of concern due to their large inter-subject and inter-site variability. We have improved existing imaging metrics of LA, WA% and AirT% with new normalization schemes (9, 10). We have added deformational variables (8, 26) using image registration (inspiration vs. expiration) techniques (27, 28). The registration-based variables have been demonstrated to be sensitive metrics when differentiating severe asthma from healthy subjects (8). While selected imaging variables have been found to differentiate asthma populations (1, 4, 8-10), a comprehensive approach that utilizes multiscale imaging-based variables to identify clinically meaningful asthma clusters has been lacking. In this study, we sought to establish a comprehensive imaging-based clustering analysis. In addition, we explored the link between structure and function captured by QCT and the commonly clinical metrics indicating lung pathophysiology to aid the development and testing of future therapeutic intervention.

## METHODS

Detailed methods related to QCT imaging and derivation of imaging-based structural and functional metrics are available in the **SUPPLEMENTARY METHODS** section.

### Human Subject Data Sets

We employed 248 subjects including 106 nonsevere and 142 severe asthmatics (**Table 1**) to derive asthma clusters, and 61 healthy subjects for the comparison with the derived clusters. The asthmatics were obtained from three different imaging centers associated with a National Institutes of Health (NIH)-sponsored multi-center study of SARP, i.e. University of Pittsburgh (PITT), Washington University in St. Louis (WSL), and University of Wisconsin (WIS). The current smokers were excluded using the inclusion criteria of “no smoking within the past 5 years and < 5 pack-years of smoking”. The severe asthmatics met the American Thoracic Society (ATS) definition of severe asthma (29, 30). The associated human studies, along with imaging protocols and informed consent, were approved by the respective institutional review boards. A subset of the subjects studied; 65 nonsevere, 60 severe asthmatic and 61 healthy subjects; were previously employed for an objective comparison of airway structures (10).

### Multiscale Imaging-based Variables

Fifty-seven QCT post-processed variables (**Figure 1 and Table E1**) were employed to identify asthma clusters. All images were analyzed utilizing the Apollo Workstation (31) and a mass-preserving image registration (28). At the segmental level, four local structural variables, i.e. bifurcation angle ( $\theta$ ), airway circularity ( $Cr$ ), wall thickness (WT) and hydraulic diameter ( $D_h$ ), were extracted at total lung capacity (TLC) scans. Each metric identifies the alteration of airway skeletal structure, heterogeneous airway luminal shape, wall thickening and luminal narrowing, respectively. Among the structural variables, WT and  $D_h$  were normalized by predicted tracheal diameter and wall thickness from healthy subjects, denoted by WT\* and  $D_h^*$ , respectively (10). This approach eliminated inter-subject variability due to gender, age and

height. Full names of each region were described in *Abbreviations used*.  $Cr$ ,  $WT^*$  and  $D_h^*$  were taken in 10 local regions to reflect regional characteristics (**Figure 1a**), whereas  $\theta$  was taken only for the Trachea and RMB because  $\theta$  was unavailable due to different airway skeletal structure of each subject. Thus, we obtained 32 local structural variables.

In addition, we employed lobar and global variables that can differentiate asthmatics from healthy subjects (8, 9). Lobar variables included AirT% at functional residual capacity (FRC), Jacobian, anisotropic deformation index (ADI), and lobar fraction of air volume change between TLC and FRC ( $\Delta V_{air}^F$ ) (**Figure 1b-d**). These variables measured abnormal gas content at expiration (air trapping), lung volume change, preferential deformation, and regional contribution of air-volume change, respectively. Global variables further included global AirT%, apical-basal distance over ventral-dorsal distance at TLC (lung shape), the ratio of air-volume changes in upper lobes to those in middle and lower lobes between TLC and FRC ( $U/(M+L)/v$ ), global Jacobian and global ADI. Thus, we obtained 20 lobar and 5 global variables. In summary, a total of 57 variables, i.e., 32 local, 20 lobar and 5 global variables, were employed in this cluster analysis (see **Table E1** for a complete list of these variables).

## Clustering and Statistical Analyses

With the 57 imaging variables, a principal component analysis (PCA) was performed to obtain a set of linearly uncorrelated variables (32, 33), so-called principal components. Since the original imaging variables were highly correlated, PCA was effective in reducing the dimensionality of the data. An optimal number of principal components was determined using the parallel analysis (**Figure E1**). We then performed clustering analysis with the principal components. We compared three general clustering methods such as *K-means*, Hierarchical and Gaussian finite mixture model-based (34) methods to find the best clustering method along with an optimal number of clusters suitable for the current imaging data (**Figure E2**), based on the internal property and stability of clusters (35, 36).

We then explored the clustering results in regards to associations with clinical metrics to see if the current clustering provides clinically meaningful results. Clinical metrics included asthma severity, gender, body mass index (BMI), onset age of asthma, pre- and post-bronchodilator pulmonary function tests (PFT), asthma quality of life questionnaire (AQLQ), asthma control questionnaire (ACQ), asthma control test (ACT), medication and exacerbation histories, atopy, bronchial hyper-responsiveness (PC<sub>20</sub>), fraction of exhaled nitric oxide (FeNO), inflammatory markers in sputum, blood and bronchial alveolar lavage (BAL). Some of clinical metrics such as sputum, BAL and PC<sub>20</sub> were only partially available. Kruskal-Wallis and chi-square tests (37) were performed to compare differences of continuous and categorical variables, respectively. We performed a total of 76 comparison tests, and  $P=0.05$  was taken as the significant level in all tests. This controls the false discovery rate (FDR) of multiple comparisons at 6.5% (e.g., 1.6% for  $P=0.01$ , and 5% for  $P=0.036$ ), estimated by the method of Benjamin and Hochberg (38).

## RESULTS

Principal component and clustering analyses, and an association of imaging-based clusters with “simple” clinical clusters are available in the **SUPPLEMENTARY RESULTS** section. Different clustering methods along with the number of clusters were tested (**Figure E2**), where *K-means* clustering with four clusters was an optimal choice, resulting in the sizes of 80, 59, 68 and 41 subjects, respectively (**Table 2**). The summary of each cluster is described in **Figure 2**.

### Imaging-based Characteristics

A stepwise forward variable selection (39) was used to find major variables which best describe the imaging-based clusters. Ten major variables with higher Wilk’s  $\lambda$  values (39) were presented to explain structural and functional alterations associated with each cluster (**Table 2**). Results of 61 healthy subjects (10) were presented to compare with those of the asthma clusters. First, airway structural variables of  $WT^*$  and  $D_h^*$  in Cluster 1 were very close to those of healthy subjects, whereas lung deformational metrics of Jacobian and ADI were relatively increased along with the small  $AirT\%$  (Total). Cluster 2 demonstrated a significant decrease of  $D_h^*$  (sLLL, BronInt and RMB) without any increase of  $WT^*$  (sRUL and sRML). This cluster demonstrated significant decreases of Jacobian (Total and LUL) and ADI (LUL). Cluster 3 demonstrated a significant increase of  $WT^*$  (sRUL and sRML), without any decrease of  $D_h^*$  (sLLL, BronInt and RMB). Cluster 4 demonstrated significant decrease of  $D_h^*$  (sLLL and RMB), increase of  $AirT\%$  (Total) and decrease of Jacobian (Total and LUL) and ADI (LUL). Thus, Cluster 1 was classified as patients with relatively normal airway structures and elevated lung deformation; Cluster 2 was classified as luminal narrowing-dominant patients ( $D_h^*\downarrow$ ) with reduced lung deformation (Jacobian $\downarrow$  and ADI $\downarrow$ ); Cluster 3 was classified as wall thickening-dominant patients ( $WT^*\uparrow$ ); Cluster 4 was classified into luminal narrowing-dominant patients ( $D_h^*\downarrow$ ) along with significant elevation of air-trapping ( $AirT\%\uparrow$ ) and decrease of lung deformation (Jacobian $\downarrow$  and ADI $\downarrow$ ).

Using the 10 major imaging variables, we further performed a decision tree analysis to detect which of the major variables best describe these imaging-based clusters in using limited imaging data. The imaging-based clusters using 57 imaging variables were predicted with an accuracy of 87% (**Figure 3**) with a complexity criterion of 0.02 (40), when using only four major variables such as Jacobian (Total), AirT% (Total),  $D_h^*$  (sLLL) and WT\* (BronInt). Note that Jacobian and AirT% were global variables, and  $D_h^*$  and WT\* were local variables. The derived clusters using the yes/no patterns in **Figure 3** are referred to as “simple” imaging-based clusters.

### **Demography, PFT and Questionnaires**

Cluster 1, with normal airway structures and increased lung deformation, was primarily populated by younger and early onset nonsevere asthmatics (**Table 3**). The cluster shows higher AQLQ scores associated with symptoms, activities and emotions. Note that higher ACQ and lower ACT scores indicate the difficulty of asthma control. Based upon the ACQ and ACT scores, asthmatics in Cluster 1 are likely to be easily controlled. Unlike Cluster 1, Cluster 2 was populated with a mix of nonsevere and severe asthmatics with low AQLQ and high ACQ-7 scores. Subjects in both Clusters 3 and 4 were mostly severe asthmatics, but they were differentiated into unique clinical characteristics. Subjects in Cluster 3 were obese (BMI $\uparrow$ ), largely females (76%) with small AQLQ scores in the area of symptoms and emotions. Cluster 4 was primarily composed of older males (51%) and late onset asthma, showing the smallest AQLQ in the area of emotional well-being and highest ACQ-7 score considering FEV<sub>1</sub> among the four clusters. ACT score was uniformly decreased in Clusters 2-4. In addition, pre-bronchodilator FEV<sub>1</sub> and forced vital capacity (FVC) % predicted values associated with Clusters 1 and 3 were somewhat smaller than normal ranges, but post-bronchodilator FEV<sub>1</sub> and FVC % predicted values returned to normal ranges (~100%) (**Table 3**). In contrast, subjects in Clusters 2 and 4 showed significant decreases in FEV<sub>1</sub> in pre-bronchodilator PFT values that persisted in post-bronchodilation.

## Biomarkers, Medication and Exacerbation Histories

According to the sputum inflammatory test, 43% patients of Cluster 4 showed an increase of eosinophils, and 78% patients of the cluster showed an increase of neutrophils. Increased neutrophils in BAL test further supported that subjects in Cluster 4 are likely to be neutrophilic-dominant asthma. On the other hand, subjects in Cluster 2 showed the lowest values for neutrophils from three different inflammatory tests indicating marginal to no inflammation. Thus, Clusters 2 and 4 seem to demonstrate marginal and neutrophilic inflammation, respectively (**Table 4**). On the other hand, Cluster 3 showed a significant decrease of blood lymphocyte along with an increase of neutrophils. Median of PC<sub>20</sub> in the cluster is much smaller than other clusters but did not reach statistical significance possibly due to small sample size. Only 8% of subjects in Cluster 1 were on nearly continuous oral corticosteroids (OCS), whereas more than 30% of subjects in Clusters 3 and 4 were populated with a large fraction of subjects on nearly continuous OCS (**Table 5**). 22% of the subjects in Cluster 1 were on SABA without inhaled corticosteroids (ICS), indicating an intermittent asthma. The subjects in Cluster 3 were populated with individuals on high-dose ICS, Long-acting  $\beta_2$ -adrenergic agonists (LABA) and Theophylline. We counted the number of controllers including nine asthma medications such as ICS, OCS, LABA, short-acting  $\beta_2$ -adnergic agonists (SABA), Leukotriene receptor antagonist (LTRA), Theophylline, Tiotropium, Ipratropium and Omalizumab. The number of controllers in Clusters 3 and 4 was significantly greater than that of Cluster 1, indicating difficulty of asthma control and/or insensitivity of corticosteroids. The Clusters 3 and 4 also showed greater urgent care visit history and need for OCS bursts more than 3 times in the past year.

## DISCUSSION

In the present study, cluster analysis using multiscale imaging-based variables not only provided statistically stable clusters with unique structural and functional characteristics, but also demonstrated meaningful associations with clinical metrics. A subset of QCT imaging-based variables, used in the present study, distinguished asthmatics from healthy subjects in previous studies (8-10). However, heretofore they have not been used together to form clinically meaningful clusters among asthmatic subjects. These clusters may provide a practical framework for discovery and outcome assessments in targeted sub-populations. As a similar effort, early work by Gupta et al. (41) used QCT-based metrics to derive three imaging-based clusters from a small group of asthmatics (N=65). Their study considered airway luminal and wall volumes normalized by BSA, but airway sizes were found to be correlated with gender, age and height, rather than BSA (10). In this work we employed the structural variables of WT and  $D_h$  with new normalization schemes to eliminate inter-subject variations due to gender, age and height. Furthermore, the current study employed an improved air-trapping measure (9) and registration-derived variables (8, 26) to link lobar function with local bronchial structure. Thus the current work is original regarding its use of a larger dataset, refined imaging-based metrics and registration-based metrics accounting for regional lung mechanics, and with respect to its further investigation of associations with clinical metrics.

Moore et al. (2) previously derived five clinical clusters. They used baseline and maximal FEV<sub>1</sub> and onset of asthma to derive “simple” clinical clusters with 80% accuracy for predicting clinical clusters. However, their study did not utilize any imaging-based variables that might serve to distinguish structural and functional alterations. We found that the “simple” clinical clusters of Moore et al. (2) are, in fact, moderately associated with the current imaging-based clusters (**Figure E3**). The association of clinical vs. imaging clusters possibly depends on the relationship of FEV<sub>1</sub> vs. airway narrowing and air-trapping (measured by  $D_h^*$  and AirT%, respectively). Previous imaging studies (10, 21) found that  $D_h^*$  and AirT% indicating large and

small airway narrowing are significantly correlated with FEV<sub>1</sub>. Note that imaging-based Cluster 4 with small FEV<sub>1</sub> exhibited significantly decreased  $D_h^*$  and increased AirT%. Although our clusters show a moderate association with the “simple” clinical clusters of Moore et al. (2), imaging-based clusters likely offer additional complementary information to these “simple” clinical clusters. This suggests that these imaging-based clusters are providing new structural and functional information. Our study further introduces “simple” imaging-based clusters using only four major (two global and two local) variables with an 87% accuracy compared with the imaging-based clusters derived from 57 variables. The “simple” imaging clusters may be more pragmatic for developing new therapy considering they reflect a more focused subset of structural and functional alterations.

In regards to airway pathology, asthma is known as a disease characterized by luminal narrowing and wall thickening due to the airway hyperresponsiveness and chronic inflammation (42). However, luminal narrowing and wall thickening tend to be considered as a mixed alteration without distinguishing between events causing one or the other independently. For instance, WA% has been used to characterize airways in QCT images with a goal of assessing airway remodeling. However, WA and LA are inter-related, thus a measure of WA% (20) does not allow one to specifically characterize airway wall remodeling (43). We also demonstrated that WA% is significantly correlated with the normalized LA (10). Therefore, in the present study, instead of WA%, we used  $D_h$  and WT normalized by predicted tracheal diameter and wall thickness from healthy subjects (10). Consequently, we were able to separate the effects of luminal narrowing from wall thickening in each subject. This helps to identify unique characteristics within each cluster. For example, Clusters “2, 4”, “3” have demonstrated bronchial characteristics associated with “luminal narrowing-dominant” and “wall thickening-dominant”, respectively.

Subjects in Cluster 1 demonstrated airway structures close to that of healthy subjects (**Table 2**), along with larger lung deformations, i.e. reduced air trapping. Post-bronchodilator

FEV<sub>1</sub> values in Cluster 1 were close to normal ranges (**Table 3**). Subjects in the cluster are likely to be intermittent asthmatics, more easily controlled, given the asthma control scores. Thus, the addition of imaging metrics to existing criteria (29, 30) in this context may enhance our ability to discriminate better-controlled asthma from severe asthma. Clusters 2, 3 and 4 demonstrate subjects with difficult to control asthma. Subjects in Cluster 2 were characterized by a mix of nonsevere and severe asthmatics with airway narrowing and marginal inflammation. This cluster demonstrated lower neutrophilic inflammatory markers in sputum, BAL and blood tests and thinner airway walls. The predominance of airway narrowing in this group was associated with the persistence of airflow limitation even with bronchodilator administration.

Clusters 3 and 4 were dominated by severe asthmatic patients. Nonetheless, their imaging and clinical characteristics were differentiable, providing a possible means of grouping severe asthmatics into independent phenotypes. For example, while Cluster 3 demonstrated significant wall thickening, Cluster 4 demonstrated significant luminal narrowing and air-trapping as reflected in the CT-based lung deformation metrics. FEV<sub>1</sub> values in Clusters 4 tended to be lower than the other clusters and, unlike Cluster 3, less reversible (**Table 3**). Cluster 3 was enriched with obese females with increased blood neutrophils and decreased blood lymphocytes. The high-dose ICS with multiple other medications in Cluster 3 might explain the observed blood neutrophilia and lymphopenia (44). The airways of this cluster demonstrate a hyper-responsiveness based on the PC<sub>20</sub> for this cluster.

Moore et al. (7) performed a cluster analysis utilizing demography, PFT results and biomarkers derived from sputum inductions and found four clusters including the following characteristics: paucigranulocytic (A), eosinophilic predominant (B), neutrophilic predominant (C) and mixed granulocytic (D). Their subjects in Cluster D demonstrated similar characteristics with our current imaging-based Cluster 4 in that they are older males with late onset asthma and persistent airflow obstruction. Cluster D from the study of Moore et al. (7) was characterized as mixed granulocytic (neutrophilic and/or eosinophilic). Similarly, our Cluster 4 demonstrates

increases in both neutrophils and eosinophils, but is more closely aligned with neutrophilic asthma based upon the BAL results. The current imaging-based cluster analysis supports the statement that the neutrophilic asthma is more strongly associated with severe asthma (7). Provided that air-trapping is indicative of small airway diseases, the current investigation is consistent with the inter-relationship, discussed by Farah et al. (45), between small airway diseases and neutrophilic inflammation. It is also known that neutrophilic asthma may be insensitive to high-dose ICS (46) which is consistent with the medication history of Cluster 4 (**Table 5**). However, whether the increase of neutrophils is due to the high-dose ICS or chronic airway inflammation is unclear.

Despite the unique imaging-based differences for severe asthma clusters presented here, medication histories were similar (**Table 5**). This suggests that imaging-based clusters or related phenotyping of asthmatics should motivate a re-thinking of interventions better guided by tools for assessing outcomes. The addition of longitudinal changes in imaging characteristics are expected to provide further strength to the clustering of nonsevere and severe asthmatics and to provide clarity regarding the stability of the clusters vs. subject transitions between clusters. The ongoing SARP studies have a longitudinal component (47) which will allow for such further exploration of imaging-based clusters in terms of membership stability vs. membership transitions. We believe that the current strength of these clusters is not in an immediate clinical application, but rather as guidance for the development and outcomes assessment of novel new intervention strategies. For instance, one might ask whether or not steroid use in Cluster 2, where there are minimal associations with inflammatory markers, is of benefit.

The distribution of asthma severity and subject characteristics within the population studied may affect overall clustering results, because the *K-means* clustering method inherently groups into similarly sized clusters. The work here demonstrates the ability of the CT imaging approach described to identify meaningful clusters within a group of severe and nonsevere asthmatics gathered across three centers. It will be of great interest to prospectively employ this

methodology in a future group of subjects that are similarly imaged and have all of the corresponding clinical parameters for comparison. The SARP 3 network, which is following the same subjects for 3 years, should allow for testing of the predictive value of these clusters by linking the imaging clusters to outcomes of severity over this time interval. It is understood that there is ongoing concern for the use of x-ray CT in young adults. Newer scanners with improved x-ray and detector technologies as well as rapid improvements in iterative reconstruction methods (48, 49) are allowing for image acquisition down to sub-mSv levels (50), which is on the order of a pair of chest films.

In conclusion, we found four distinct clusters of asthmatic phenotypes (**Figure 2**): Cluster 1, nonsevere asthmatics with normal airway structure; Cluster 2, a mix of nonsevere and severe asthma with airway luminal narrowing without wall thickening; Cluster 3, early-onset obese female severe asthmatics with reversible lung function and airway wall thickening; Cluster 4, older males, late onset neutrophil predominant inflammation with significant air-trapping and reduction in lung deformation. The unique structural and functional alterations observed in each cluster provide new insights on the existing pathophysiology of asthma. The clustering membership employed in this analysis may prove to be useful for the development of effective interventions of asthmatics along with a future study. Furthermore, the approach presented here provides a framework whereby a growing wealth of imaging-derived metrics can be optimally utilized to identify sub-populations within many disease categories including asthma.

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## TABLES

Table 1. Demography, baseline (Prebronchodilator) and maximal (Postbronchodilator) pulmonary function tests for 106 nonsevere asthmatics, 142 severe asthmatics, and 61 healthy subjects

|                                             | Nonsevere<br>asthmatics | Severe<br>asthmatics   | Healthy<br>subjects  | P value  |
|---------------------------------------------|-------------------------|------------------------|----------------------|----------|
| Demography                                  | N = 106                 | N = 142                | N = 61               |          |
| Age, yrs                                    | 36.0 (12.2)             | 46.9 (13.1)            | 35.6 (14.2)          | < 0.0001 |
| BMI, kg/m <sup>2</sup>                      | 29.6 (7.6)              | 33.6 (8.4)             | 26.6 (6.3)           | < 0.0001 |
| Asthma duration, yrs                        | 21.6 (12.8)             | 28.7 (15.3)            | -                    | < 0.0005 |
| Gender, Female                              | 68 (64%)                | 90 (63%)               | 36 (59%)             | = 0.788  |
| Race, Caucasian/ African<br>American/ Other | 78/20/8<br>(74/19/7%)   | 94/37/11<br>(66/26/8%) | 50/3/8<br>(82/5/13%) | = 0.011  |
| Baseline lung function <sup>†</sup>         | N = 106                 | N = 142                | N = 61               |          |
| FEV <sub>1</sub> % predicted                | 85 (17)                 | 65 (22)                | 101 (13)             | < 0.0001 |
| FVC % predicted                             | 94 (14)                 | 77 (17)                | 100 (12)             | < 0.0001 |
| FEV <sub>1</sub> /FVC × 100                 | 74 (10)                 | 66 (12)                | 83 (6)               | < 0.0001 |
| Maximal lung function <sup>‡</sup>          | N = 102                 | N = 141                | N = 36               |          |
| FEV <sub>1</sub> % predicted                | 96 (14)                 | 79 (20)                | 103 (9)              | < 0.0001 |
| FVC % predicted                             | 100 (13)                | 90 (16)                | 101 (9)              | < 0.0001 |
| FEV <sub>1</sub> /FVC × 100                 | 79 (9)                  | 70 (11)                | 85 (6)               | < 0.0001 |

Values expressed as mean (SD) or number (%). Kruskal-Wallis and chi-square tests were performed for continuous and categorical variables. <sup>†</sup>Baseline (Prebronchodilator) values with greater than six hours withhold of bronchodilators. <sup>‡</sup>Maximal (Postbronchodilator) values after six to eight puffs of albuterol.

Table 2. Major structural and functional imaging-based variables in four imaging-based clusters and healthy subjects

| Variable           | Region  | Wilk's $\lambda$ value | Cluster 1<br>(N = 80) | Cluster 2<br>(N = 59) | Cluster 3<br>(N = 68) | Cluster 4<br>(N = 41) | P value  | Healthy subjects<br>(N = 61) |
|--------------------|---------|------------------------|-----------------------|-----------------------|-----------------------|-----------------------|----------|------------------------------|
| Jacobian           | Total   | 0.317                  | 2.48<br>(0.33)        | 1.69<br>(0.26)        | 1.91<br>(0.22)        | 1.47<br>(0.17)        | < 0.0001 | 2.05<br>(0.44)               |
| AirT%              | Total   | 0.177                  | 5.8<br>(5.5)          | 8.6<br>(7.0)          | 8.9<br>(6.3)          | 29.7<br>(11.6)        | < 0.0001 | 7.8<br>(8.0)                 |
| $D_h^*$            | sLLL    | 0.123                  | 0.338<br>(0.038)      | 0.269<br>(0.044)      | 0.344<br>(0.044)      | 0.301<br>(0.046)      | < 0.0001 | 0.338<br>(0.041)             |
| WT*                | sRUL    | 0.098                  | 0.609<br>(0.037)      | 0.587<br>(0.043)      | 0.646<br>(0.040)      | 0.607<br>(0.039)      | < 0.0001 | 0.612<br>(0.049)             |
| $D_h^*$            | BronInt | 0.088                  | 0.649<br>(0.047)      | 0.586<br>(0.055)      | 0.670<br>(0.054)      | 0.609<br>(0.052)      | < 0.0001 | 0.622<br>(0.049)             |
| $\Delta V_{air}^F$ | RUL     | 0.080                  | 0.178<br>(0.025)      | 0.197<br>(0.041)      | 0.190<br>(0.027)      | 0.172<br>(0.034)      | < 0.0005 | 0.161<br>(0.020)             |
| Jacobian           | LUL     | 0.075                  | 2.44<br>(0.38)        | 1.69<br>(0.26)        | 1.88<br>(0.26)        | 1.42<br>(0.18)        | < 0.0001 | 1.94<br>(0.47)               |
| $D_h^*$            | RMB     | 0.071                  | 0.843<br>(0.074)      | 0.755<br>(0.074)      | 0.870<br>(0.074)      | 0.796<br>(0.068)      | < 0.0001 | 0.828<br>(0.062)             |
| WT*                | sRML    | 0.068                  | 0.581<br>(0.047)      | 0.531<br>(0.049)      | 0.602<br>(0.039)      | 0.576<br>(0.040)      | < 0.0001 | 0.589<br>(0.056)             |
| ADI                | LUL     | 0.065                  | 0.487<br>(0.080)      | 0.321<br>(0.092)      | 0.382<br>(0.085)      | 0.247<br>(0.066)      | < 0.0001 | 0.352<br>(0.103)             |

Values expressed as mean (SD). The major imaging-based variables were selected by Wilk's  $\lambda$  value of a stepwise forward variable selection method. Kruskal-Wallis tests were performed. Full names of each variable or region were described in *Abbreviations used*. The ten major variables are composed of two global [Jacobian (Total) and AirT% (Total)], three lobar [ $\Delta V_{air}^F$ , Jacobian (LUL) and ADI (LUL)], and five local variables [ $D_h^*$  (sLLL), WT\* (sRUL),  $D_h^*$  (BronInt),  $D_h^*$  (RMB) and WT\* (sRML)].

Table 3. Demography, baseline (Pre-bronchodilator) and maximal (Post-bronchodilator) PFTs, AQLQ, ACQ and ACT scores in four imaging-based clusters

|                                                                          | Cluster 1            | Cluster 2            | Cluster 3            | Cluster 4           | P value  |
|--------------------------------------------------------------------------|----------------------|----------------------|----------------------|---------------------|----------|
| <i>Demography</i>                                                        |                      |                      |                      |                     |          |
|                                                                          | N = 80               | N = 59               | N = 68               | N = 41              |          |
| Asthma severity (Severe asthma, %)                                       | 32 (40%)             | 30 (51%)             | 50 (74%)             | 30 (73%)            | < 0.0001 |
| Gender (Female, %)                                                       | 51 (64%)             | 35 (59%)             | 52 (76%)             | 20 (49%)            | = 0.026  |
| Race, No. (Caucasian/African American/Other)                             | 60/15/5 (75/19/6%)   | 37/16/6 (63/27/10%)  | 45/17/6 (66/25/9%)   | 30/9/2 (73/22/5%)   | = 0.763  |
| Age (yrs)                                                                | 37.8 (13.6)          | 40.9 (13.7)          | 45.0 (13.3)          | 48.4 (12.0)         | < 0.0005 |
| BMI (kg/m <sup>2</sup> )                                                 | 31.1 (7.8)           | 30.1 (7.5)           | 35.1 (8.8)           | 30.7 (8.1)          | < 0.005  |
| Asthma duration (yrs)                                                    | 24.1 (14.2)          | 26.3 (14.0)          | 28.0 (16.0)          | 24.1 (14.2)         | = 0.380  |
| Onset of asthma (yrs)                                                    | 13.8 (13.7)          | 14.6 (15.2)          | 17.0 (16.0)          | 24.3 (17.5)         | = 0.015  |
| Number of < 12 yrs, Onset of asthma (%)                                  | 51 (64%)             | 35 (59%)             | 35 (51%)             | 13 (32%)            | < 0.01   |
| Center, No. (PITT/WSL/WIS)                                               | 22/25/33 (28/31/41%) | 20/17/22 (34/29/37%) | 23/21/24 (34/31/35%) | 16/17/8 (39/41/20%) | = 0.384  |
| <i>Asthma quality of life questionnaire (AQLQ)</i>                       |                      |                      |                      |                     |          |
|                                                                          | N = 80               | N = 58               | N = 68               | N = 41              |          |
| Symptoms                                                                 | 5.22 (1.2)           | 4.59 (1.5)           | 4.37 (1.5)           | 4.32 (1.4)          | < 0.001  |
| Activities                                                               | 5.38 (1.4)           | 4.55 (1.5)           | 4.62 (1.4)           | 4.49 (1.3)          | < 0.0005 |
| Emotions                                                                 | 5.10 (1.6)           | 4.42 (1.8)           | 4.35 (1.8)           | 4.16 (1.7)          | < 0.01   |
| Environment                                                              | 4.95 (1.5)           | 4.63 (1.7)           | 4.66 (1.4)           | 4.51 (1.4)          | = 0.252  |
| <i>Asthma control questionnaire (ACQ) and Asthma control test (ACT)*</i> |                      |                      |                      |                     |          |
|                                                                          | N = 40               | N = 25               | N = 35               | N = 23              |          |
| ACQ-6 score (without FEV <sub>1</sub> )                                  | 1.14 (0.9)           | 1.79 (1.4)           | 1.90 (1.3)           | 2.17 (1.4)          | < 0.01   |
| ACQ-7 score (with FEV <sub>1</sub> )                                     | 1.27 (0.8)           | 2.14 (1.3)           | 2.03 (1.3)           | 2.49 (1.3)          | < 0.001  |

|                                     |            |            |            |            |          |
|-------------------------------------|------------|------------|------------|------------|----------|
| ACT score                           | 19.4 (4.1) | 14.9 (4.9) | 14.9 (4.9) | 14.7 (5.2) | < 0.0001 |
| <i>PFT Baseline lung function †</i> |            |            |            |            |          |
|                                     | N = 80     | N = 59     | N = 68     | N = 41     |          |
| FEV1 % predicted                    | 83 (18)    | 69 (21)    | 75 (22)    | 58 (21)    | < 0.0001 |
| FVC % predicted                     | 93 (15)    | 80 (19)    | 83 (17)    | 75 (17)    | < 0.0001 |
| FEV1/FVC                            | 73 (10)    | 69 (11)    | 71 (11)    | 60 (13)    | < 0.0001 |
| <i>PFT Maximal lung function ‡</i>  |            |            |            |            |          |
|                                     | N = 77     | N = 58     | N = 68     | N = 40     |          |
| FEV1 % predicted                    | 94 (16)    | 81 (20)    | 89 (18)    | 73 (18)    | < 0.0001 |
| FVC % predicted                     | 100 (15)   | 89 (16)    | 94 (15)    | 89 (16)    | < 0.001  |
| FEV1/FVC                            | 77 (9)     | 74 (12)    | 76 (9)     | 65 (12)    | < 0.0001 |

Data presented as number (%) or mean (SD). Kruskal-Wallis and chi-square tests were performed for continuous and categorical variables, respectively. \*ACQ and ACT scores were only acquired in SARP phase 3. † Prebronchodilator values with greater than six hours withhold of bronchodilators. ‡ Postbronchodilator values after six to eight puffs of albuterol. Full names of each variable were described in *Abbreviations used*.

Table 4. Characteristics of biomarkers in four imaging-based clusters

|                                | Cluster 1 |               | Cluster 2 |               | Cluster 3 |               | Cluster 4 |               | P value |
|--------------------------------|-----------|---------------|-----------|---------------|-----------|---------------|-----------|---------------|---------|
|                                | N         | Value         | N         | Value         | N         | Value         | N         | Value         |         |
| <i>Inflammatory biomarkers</i> |           |               |           |               |           |               |           |               |         |
| Sputum induction               | 45        |               | 37        |               | 42        |               | 28        |               |         |
| % Eosinophils†                 |           | 0.5 (0.0-1.6) |           | 0.5 (0.1-4.7) |           | 0.6 (0.2-2.6) |           | 1.1 (0.1-6.7) | = 0.550 |
| % Eosinophils > 2.5%           |           | 6 (13%)       |           | 10 (27%)      |           | 11 (26%)      |           | 12 (43%)      | = 0.047 |
| % Neutrophils†                 |           | 62 (42-77)    |           | 48 (24-73)    |           | 60 (42-72)    |           | 70 (55-82)    | = 0.209 |
| % Neutrophils > 50%            |           | 27 (60%)      |           | 16 (43%)      |           | 27 (64%)      |           | 22 (78%)      | = 0.034 |
| BAL                            | 42        |               | 32        |               | 33        |               | 24        |               |         |
| % Eosinophils†                 |           | 0.4 (0.0-1.0) |           | 0.0 (0.0-0.5) |           | 0.3 (0.0-1.0) |           | 0.2 (0.0-0.8) | = 0.128 |
| % Neutrophils†                 |           | 1.9 (0.7-3.6) |           | 0.7 (0.0-2.0) |           | 2.0 (1.0-5.5) |           | 2.8 (1.7-5.4) | < 0.005 |
| Blood/serum                    | 77        |               | 58        |               | 65        |               | 38        |               |         |
| Total WBC‡, N/μl               |           | 6746 (1)      |           | 6655 (1)      |           | 7615 (1)      |           | 6939 (1)      | = 0.157 |
| % Eosinophils†                 |           | 3.0 (2.0-5.0) |           | 3.0 (2.0-4.8) |           | 3.0 (1.0-4.0) |           | 3.9 (2.2-5.0) | = 0.273 |
| % Neutrophils†                 |           | 56 (53-62)    |           | 55 (48-63)    |           | 62 (55-70)    |           | 58 (51-67)    | < 0.01  |
| % Lymphocyte†                  |           | 31 (26-35)    |           | 34 (25-40)    |           | 27 (21-32)    |           | 31 (22-35)    | < 0.001 |
| <i>Additional biomarkers</i>   |           |               |           |               |           |               |           |               |         |
| Atopy status                   | 76        |               | 56        |               | 62        |               | 39        |               |         |
| ≥ 1 positive SPT               |           | 64 (84%)      |           | 50 (89%)      |           | 52 (84%)      |           | 28 (72%)      | = 0.158 |
| N of positive SPT†             |           | 4 (1.8-7.0)   |           | 5 (2.0-8.0)   |           | 4 (1.0-7.0)   |           | 3 (0-5.8)     | = 0.161 |
| Bronchial hyperresponsiveness  | 47        |               | 35        |               | 30        |               | 9         |               |         |
| PC <sub>20</sub> ‡, mg/ml      |           | 1.6 (0.5-4.5) |           | 1.7 (0.6-3.9) |           | 0.9 (0.3-1.8) |           | 4.5 (0.4-7.1) | = 0.128 |
| PC <sub>20</sub> < 1.0 mg/ml   |           | 17 (36%)      |           | 12 (34%)      |           | 19 (63%)      |           | 3 (33%)       | = 0.061 |
| Total IgE‡, IU/ml              | 73        | 103 (4)       | 56        | 118 (4)       | 60        | 101 (5)       | 35        | 113 (5)       | = 0.976 |
| FeNO‡, ppb                     | 69        | 23.6 (1.9)    | 48        | 25.1 (2.0)    | 54        | 28.5 (2.4)    | 38        | 26.8 (2.5)    | = 0.345 |

Full names of each variable were described in *Abbreviations used*. † Data expressed as median (interquartile range). ‡ Data expressed as geometric mean (geometric SD). Kruskal-Wallis and chi-square tests were performed for continuous and categorical variables, respectively.

Table 5. Medication use and exacerbation history in four imaging-based clusters

|                                                   | Cluster 1 | Cluster 2 | Cluster 3 | Cluster 4 | P value* |
|---------------------------------------------------|-----------|-----------|-----------|-----------|----------|
| <i>Medication use</i>                             |           |           |           |           |          |
|                                                   | N = 80    | N = 59    | N = 68    | N = 41    |          |
| OCS $\geq$ 50% of year                            | 6 (8%)    | 12 (20%)  | 24 (35%)  | 15 (37%)  | < 0.0001 |
| High-dose ICS †                                   | 35 (44%)  | 30 (51%)  | 52 (76%)  | 27 (66%)  | < 0.0005 |
|                                                   | N = 78    | N = 57    | N = 65    | N = 40    |          |
| SABA without use of ICS, LABA and LTRA            | 17 (22%)  | 7 (12%)   | 3 (5%)    | 3 (8%)    | = 0.013  |
| ICS                                               | 60 (77%)  | 48 (84%)  | 61 (94%)  | 35 (88%)  | = 0.042  |
| ICS + LABA                                        | 50 (64%)  | 42 (74%)  | 55 (85%)  | 31 (78%)  | = 0.045  |
| ICS + LABA + LTRA                                 | 18 (23%)  | 20 (35%)  | 23 (35%)  | 11 (28%)  | = 0.321  |
| ICS + LABA + OCS                                  | 9 (12%)   | 11 (19%)  | 20 (31%)  | 13 (33%)  | = 0.014  |
| Theophylline                                      | 0 (0%)    | 8 (14%)   | 10 (15%)  | 2 (5%)    | < 0.005  |
| Anti-IgE (Omalizumab)                             | 5 (6%)    | 7 (12%)   | 12 (18%)  | 9 (23%)   | = 0.059  |
| Number of medications ‡                           | 3.1 (1.6) | 3.6 (1.7) | 4.2 (1.6) | 4.1 (1.3) | < 0.0005 |
| <i>Exacerbation History</i>                       |           |           |           |           |          |
|                                                   | N = 80    | N = 59    | N = 68    | N = 41    |          |
| $\geq$ 1 urgent care visits / year                | 19 (24%)  | 19 (32%)  | 36 (53%)  | 21 (51%)  | < 0.001  |
| $\geq$ 3 OCS, bursts / year                       | 24 (30%)  | 24 (41%)  | 37 (54%)  | 22 (54%)  | = 0.011  |
| Deterioration with a reduction in OCS or ICS dose | 25 (31%)  | 26 (44%)  | 30 (44%)  | 20 (49%)  | = 0.197  |
|                                                   | N = 78    | N = 57    | N = 65    | N = 40    |          |
| ICU Hospitalized in the past year                 | 1 (1%)    | 2 (4%)    | 5 (8%)    | 0 (0%)    | = 0.101  |
| Ever needed a ventilator                          | 2 (3%)    | 8 (14%)   | 7 (11%)   | 5 (13%)   | = 0.091  |

Full names of each variable were described in *Abbreviations used*. Data presented as number of subject (%) or mean (SD). \* Kruskal-Wallis and chi-square tests were performed for continuous and categorical variables, respectively. † High-dose ICS dose equivalent to 1,000 or greater fluticasone propionate daily. ‡ Counts of nine medications: ICS, OCS, LABA, SABA, LTRA, Theophylline, Tiotropium, Ipratropium and Omalizumab.

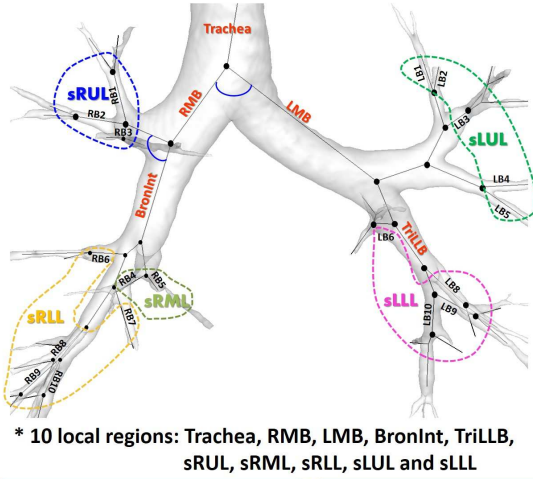
**FIGURE CAPTIONS**

Figure 1. Multiscale imaging-based variables employed for the cluster analysis. Labels of ten selected local regions are shown in (a). Full names of each variable or region are described in *Abbreviations used*.

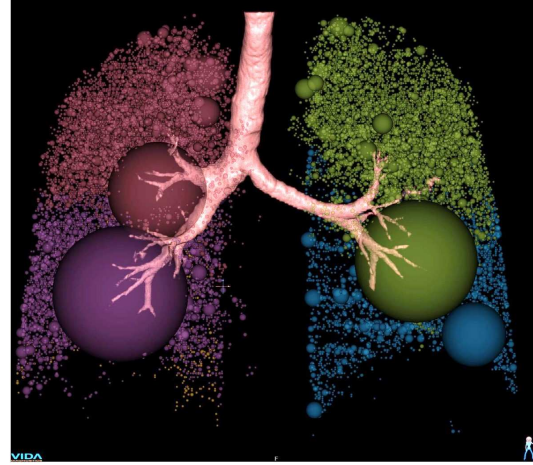
Figure 2. A summary of respective imaging clusters. Note that most of the severe asthmatics were populated in Clusters 3 and 4.

Figure 3. A “simple” scheme for predicting image-based clusters using only four major variables that has 87% accuracy compared with “original” imaging-based clusters using fifty seven variables

**A. Inspiration image-based local structures:**  
 $\theta$ ,  $Cr$ ,  $WT^*$ , and  $D_n^*$

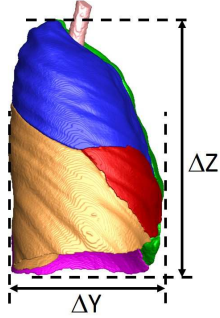


**B. Expiration image-based global and lobar function: AirT%**



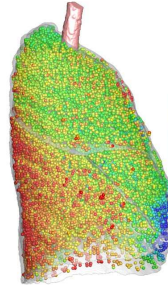
**C. Global structure:**

Lung shape =  $\Delta Z / \Delta Y$

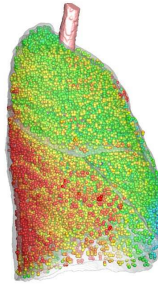


**D. Registration-based global and lobar functions:**

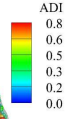
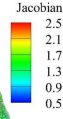
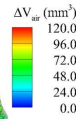
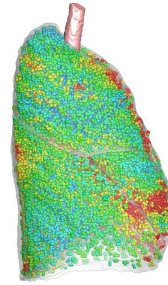
$U/(M+L)|v$ , and  $\Delta V_{air}^F$



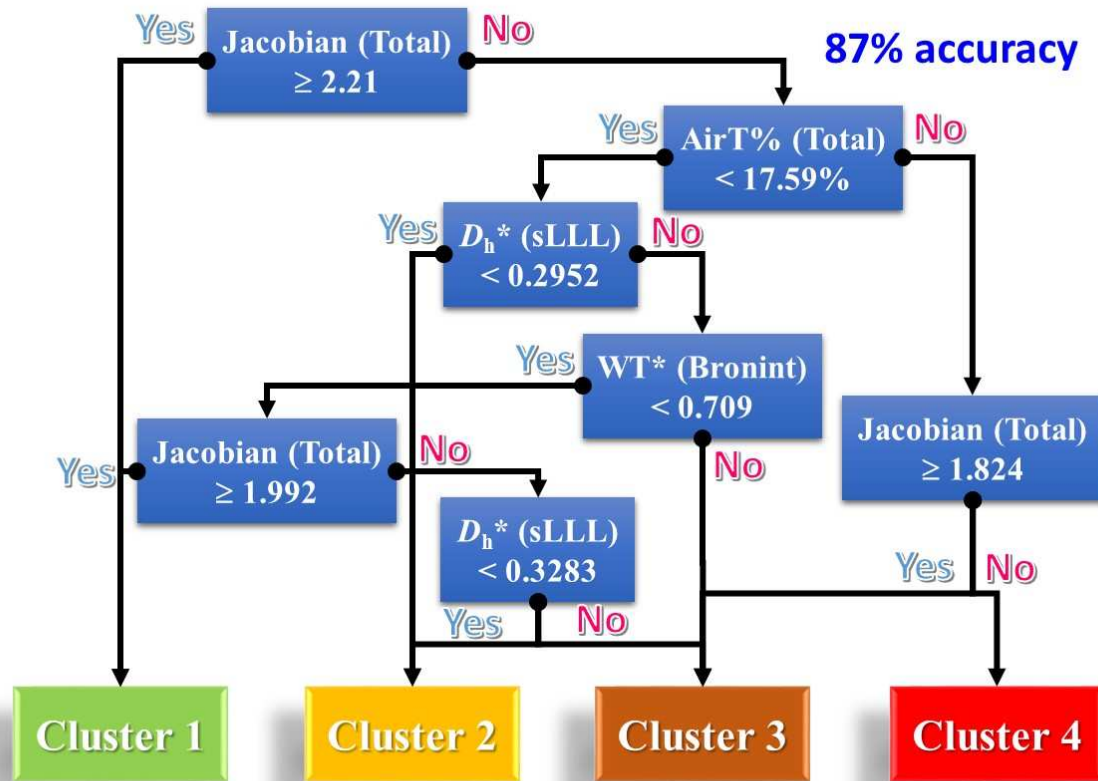
Jacobian



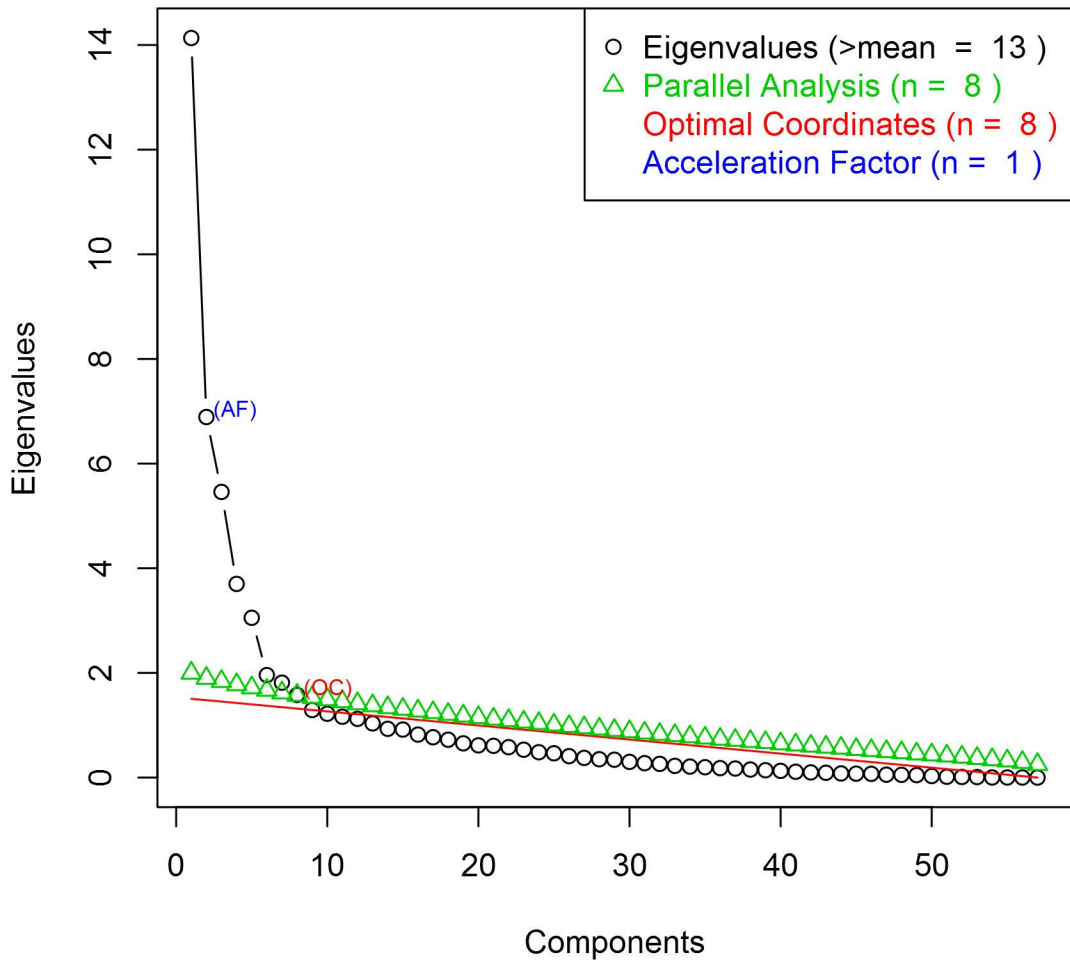
ADI



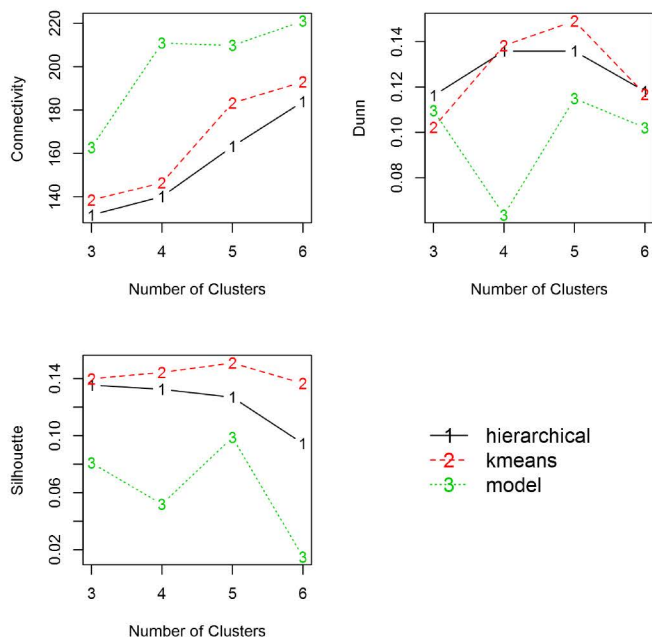
|                  | Imaging characteristics                                                                                                                                                                                                                                                     | Clinical characteristics                                                                                                                                                                                                                                  |
|------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <b>Cluster 1</b> | <ul style="list-style-type: none"> <li>• Normal airway structure</li> <li>• Increased lung deformation (Jacobian and ADI<math>\uparrow</math>)</li> </ul>                                                                                                                   | <ul style="list-style-type: none"> <li>• Younger, early onset</li> <li>• Nonsevere asthma</li> <li>• Reversible lung function</li> <li>• Easy to control asthma symptoms</li> </ul>                                                                       |
| <b>Cluster 2</b> | <ul style="list-style-type: none"> <li>• Airway luminal narrowing (<math>D_h^*\downarrow</math>)</li> <li>• No airway wall thickening (WT*)</li> <li>• Significant reduction of lung deformation (Jacobian and ADI<math>\downarrow</math>)</li> </ul>                       | <ul style="list-style-type: none"> <li>• Nonsevere and severe asthma</li> <li>• Persistently altered lung function</li> <li>• Marginal to no inflammation</li> <li>• Difficult to control asthma symptoms</li> </ul>                                      |
| <b>Cluster 3</b> | <ul style="list-style-type: none"> <li>• Airway wall thickening (WT*<math>\uparrow</math>)</li> <li>• No airway luminal narrowing (<math>D_h^*</math>)</li> <li>• Moderate reduction of lung deformation (Jacobian and ADI<math>\downarrow</math>)</li> </ul>               | <ul style="list-style-type: none"> <li>• Obese, female-dominant</li> <li>• Severe asthma</li> <li>• Reversible lung function</li> <li>• Blood lymphopenia</li> <li>• Difficult to control asthma symptoms</li> </ul>                                      |
| <b>Cluster 4</b> | <ul style="list-style-type: none"> <li>• Airway luminal narrowing (<math>D_h^*\downarrow</math>)</li> <li>• Significant reduction of lung deformation (Jacobian and ADI<math>\downarrow</math>)</li> <li>• Significant air-trapping (AirT%<math>\uparrow</math>)</li> </ul> | <ul style="list-style-type: none"> <li>• Older, late onset, male-dominant</li> <li>• Severe asthma</li> <li>• Persistently altered lung function</li> <li>• Neutrophilic-dominant inflammation</li> <li>• Difficult to control asthma symptoms</li> </ul> |



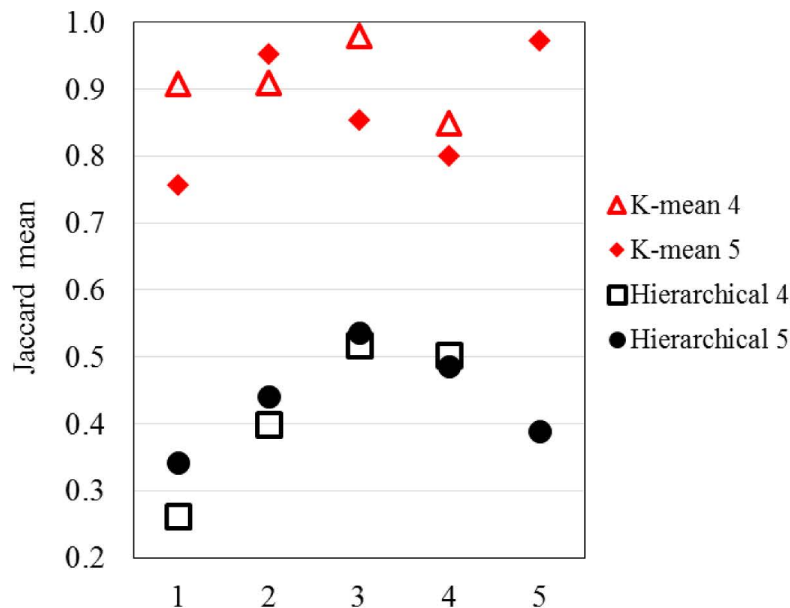
| “Simple”<br>Imaging clusters | Imaging-based clusters |    |    |    |
|------------------------------|------------------------|----|----|----|
|                              | 1                      | 2  | 3  | 4  |
| 1                            | 74                     | 0  | 5  | 0  |
| 2                            | 0                      | 46 | 5  | 1  |
| 3                            | 6                      | 8  | 58 | 3  |
| 4                            | 0                      | 5  | 0  | 37 |



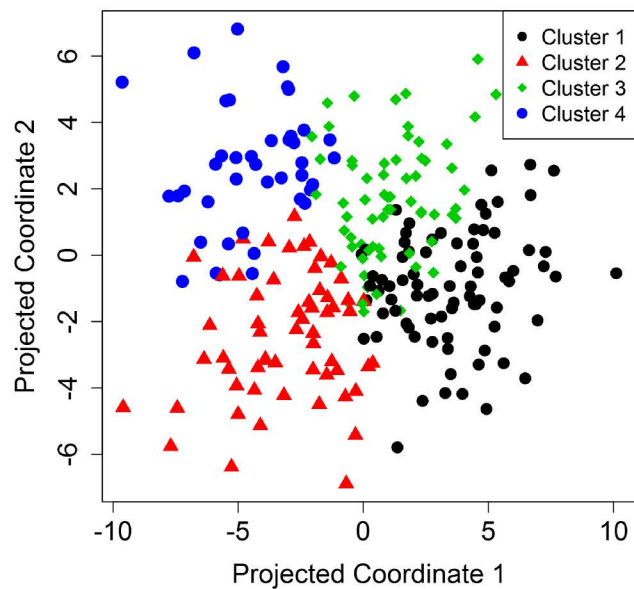
## a. Internal property



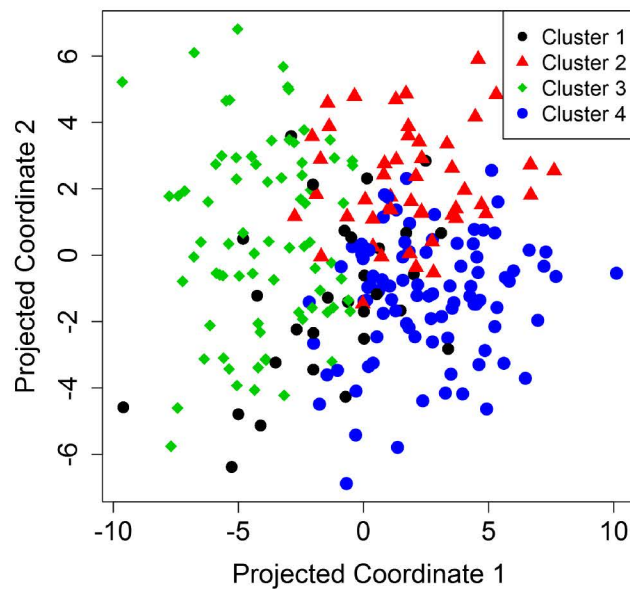
## b. Clustering stability



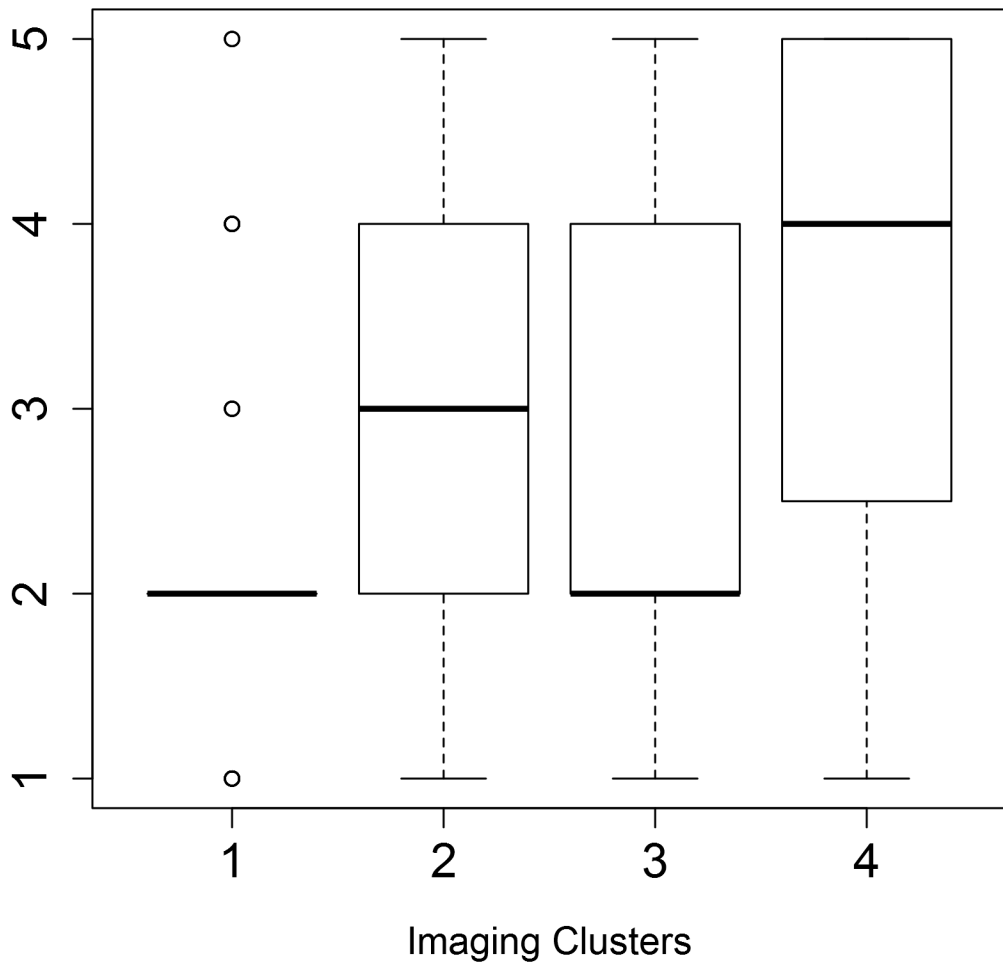
## c. K-means clustering



## d. Hierarchical clustering



Simple Clinical Clusters



## SUPPLEMENTARY METHODS

### Quantitative Computed Tomography (QCT) Imaging

In Severe Asthma Research Program (SARP) phases 1 and 2, scanners and imaging protocols of three centers were outlined in Choi et al. (E1). Imaging within SARP phase 3 followed the imaging protocols established by SPIROMICS (E2) with the exception that expiratory scans were carried out at functional residual capacity (FRC) rather than residual volume (RV) for consistency with earlier SARP data. The protocol called for several deep inspiratory maneuvers prior to coaching to total lung capacity (TLC) and FRC to provide a standardized lung volume history. There have been several reports associated with the effects of deep inspiration on airway dimension in asthmatics (E3, 4). However, the current imaging protocol is a generally accepted approach to standardize parenchymal expansion not only in SARP but also in other multicenter studies including the Multi-Ethnic Study of Atherosclerosis (MESA) (E5), the SubPopulations and InteRmediate Outcome Measures In Chronic obstructive pulmonary disease Study (SPIROMICS) (E2) and Genetic Epidemiology of COPD (COPDGene) (E6). We applied deep inspiration instructions prior to scanning of all subjects, and the interpretation of results should include an understanding that airway metrics were made following deep inspirations.

### Post-processing of QCT Images

Post-processing was done with Apollo Workstation (E7) and a mass-preserving image registration method (E8). The Apollo Workstation guarantees the segmentation accuracy within the voxel size (E7). The Apollo Workstation has been used in many large multi-center studies, demonstrating the robust lung and airway segmentations and repeatability across users (E9, 10). In order to minimize the effects of QCT resolution and segmentation software, we limited our interest of regions of airway tree up to tertiary bronchi, i.e., segmental bronchi, generation number of  $\sim 3^{\text{rd}}\text{-}5^{\text{th}}$ , which typically have diameters of 4–6 mm ( $\geq 2\text{mm}$ ). The more distal airway

pathophysiology was indexed by use of our measure of air trapping on the expiratory scan. Since the resolution of QCT voxel was less than 1 mm, the segmented airways should not be affected by the use of different Workstations if the Workstation guarantees reliable accuracy. In addition, the image registration method (E8) was validated with the displacement of landmarks of the vessel trees in the large deformation.

### Derivation of Segmental Structural Variables

At the segmental level, four local structural variables, i.e. bifurcation angle ( $\theta$ ), airway circularity ( $Cr$ ), wall thickness (WT) and hydraulic diameter ( $D_h$ ), were extracted at total lung capacity (TLC) scans. The Apollo Workstation provided 1-D skeletal structure, average luminal area (LA), total area (TA), and perimeter of LA ( $P_e$ ). The LA, TA and  $P_e$  were averaged from the middle region (30-70%) of an airway segment. Using the 1-D airway skeleton, the  $\theta$  was calculated as

$$\theta = \cos^{-1} \left( \frac{\mathbf{d}_1 \cdot \mathbf{d}_2}{|\mathbf{d}_1| |\mathbf{d}_2|} \right), \quad (1)$$

where  $\mathbf{d}_1$  and  $\mathbf{d}_2$  are directional vectors of daughter branches of a named segment, respectively. Average luminal diameter ( $D_{ave}$ ) and outer diameter ( $D_{outer}$ ) were calculated as  $\sqrt{4LA/\pi}$  and  $\sqrt{4TA/\pi}$ , respectively. Given the derived metrics,  $Cr$ , WT and  $D_h$  were calculated as follows:

$$Cr = \frac{\pi D_{ave}}{P_e}, \quad (2)$$

$$WT = D_{outer} - D_{ave}, \quad (3)$$

$$D_h = \frac{4LA}{P_e}. \quad (4)$$

Among the four structural variables, WT and  $D_h$  were normalized by predicted tracheal diameter and wall thickness from healthy subjects, denoted by  $WT^*$  and  $D_h^*$ , respectively (E1). The predicted tracheal diameter and wall thickness were derived as follows:

$$D_{trachea, pred} \text{ (mm)} = 16.446 - 2.4019 \text{ gender} - 0.298809 \text{ gender} \cdot \text{age} + 0.0284836 \text{ age} \cdot \text{height} + 0.1786604 \text{ gender} \cdot \text{age} \cdot \text{height}, \quad (5)$$

$$WT_{\text{trachea,pred}} \text{ (mm)} = 4.5493 - 0.5007 \text{ gender} + 0.3007 \log_{10}(\text{age}) \cdot \text{height}, \quad (6)$$

where gender, age and height take the value/unit of 0 (male) or 1 (female), years, and meter, respectively. Note that the predicted tracheal diameter and wall thickness were significantly correlated with the gender, age and height, so that they effectively eliminated the inter-subject variability (E1), unlike the existing body surface area (BSA)-based normalization (E11).  $Cr$ ,  $WT$  and  $D_h$  were taken in ten local regions to reflect regional characteristics (**Figure 1a**), i.e. Trachea, right main bronchus (RMB), left main bronchus (LMB), intermediate bronchus (BronInt), trifurcation left lower bronchus (TriLLB), sub-grouped left upper lobe [sLUL: left bronchus (LB1-5)], sub-grouped left lower lobe (sLLL: LB6, 8-10), sub-grouped right upper lobe [sRUL: right bronchus (RB1-3)], sub-grouped right middle lobe (sRML: RB4-5) and sub-grouped right lower lobe (sRLL: RB6-10). On the other hand,  $\theta$  was taken only for the Trachea and RMB, because  $\theta$  was unavailable due to different airway skeletal structure of each subject.

### Derivation of Global/Lobar Functional Variables

As for AirT%, since the density-threshold-based air-trapping method, e.g. CT density ( $I$ ) of  $-850$  HU, was sensitive to the imaging protocols (E12), we employed a fraction-threshold-based air-trapping method using a fraction ( $\beta$ ) of 0.9 as follows:

$$I_{\text{threshold}} = \beta HU_{\text{air,trachea}} + (1 - \beta) HU_{\text{tissue}}, \quad (7)$$

where  $HU_{\text{air,trachea}}$  was Hounsfield Unit extracted from trachea, and  $HU_{\text{tissue}}$  was Hounsfield Unit of tissue as set to 55. Thus, each QCT scan has its own  $I_{\text{threshold}}$ , rather than having the same  $I_{\text{threshold}}$  of  $-850$ . If  $I$  was below  $I_{\text{threshold}}$  in a voxel, the voxel was regarded as air-trapped. The AirT% was calculated as the ratio of the number of air-trapped voxels to the number of voxels in the whole lung (or respective lobes) at FRC. The approach successfully eliminated the inter-site and inter-subject variations.

In addition, in order to obtain deformation variables, we used a mass-preserving image registration, providing a warping function [ $\mathbf{T}(\mathbf{x})$ ] matching a local position  $\mathbf{x}$  of reference image

to the corresponding position of the floating image. Thus local air volume change  $[\Delta V_{air}(\mathbf{x})]$  was computed as follows:

$$\Delta V_{air}(\mathbf{x}) = V_{air}^{ref}(\mathbf{x}) - V_{air}^f(\mathbf{T}(\mathbf{x})), \quad (8)$$

where  $V_{air}^{ref}(\mathbf{x})$  and  $V_{air}^f(\mathbf{T}(\mathbf{x}))$  were local air volume at reference and corresponding one at floating images, respectively. The sum of  $\Delta V_{air}(\mathbf{x})$  was calculated in lobes and whole lung, and then lobar fraction of air volume change ( $\Delta V_{air}^F$ ) was computed as the ratio of  $[\sum_{\mathbf{x} \in Lobe} \Delta V_{air}(\mathbf{x})]$  to  $[\sum_{\mathbf{x} \in whole\ lung} \Delta V_{air}(\mathbf{x})]$ . Similarly, the global metric of U/(M+L)|v can be calculated as the ratio of  $[\sum_{\mathbf{x} \in RUL, LUL} \Delta V_{air}(\mathbf{x})]$  to  $[\sum_{\mathbf{x} \in RML, RLL, LLL} \Delta V_{air}(\mathbf{x})]$ .

Furthermore, using the concept of deformation gradient tensor (E13), eigenvalues ( $\lambda_1 > \lambda_2 > \lambda_3 > 0$ ) for each local volume were derived. Jacobian and ADI were then calculated as follows:

$$\text{Jacobian} = \lambda_1 \lambda_2 \lambda_3 \quad (9)$$

$$\text{ADI} = \sqrt{\left(\frac{\lambda_1 - \lambda_2}{\lambda_2}\right)^2 + \left(\frac{\lambda_2 - \lambda_3}{\lambda_3}\right)^2} \quad (10)$$

We extracted medians of Jacobian and ADI in the whole lung and each lobe to reduce a large number of data at the voxel level. The detailed methods for the fraction-threshold based air-trapping and image registration methods were described in the references (E14-16).

## SUPPLEMENTARY RESULTS

### Principal Component Analysis

A principal component analysis (PCA) was conducted to perform dimension reduction, because strong correlations exist among the original raw 57 imaging variables. Using the parallel analysis (E17, 18), we kept principal components associated with eigenvalues from the current data which were larger than 5<sup>th</sup> percentile of eigenvalues obtained from random normal data. As such, the optimal number of principal components was determined as eight (**Figure E1**)

explaining 68% of the variation in the original data (**Table E1**). Eight component loadings were reported in **Table E1**. The table shows the clustering pattern of the imaging variables in constructing the principal components. The first principal component was mainly an average effect of Jacobian and ADI; the second one was related to  $WT^*$  and  $D_h^*$  (mainly in lobar bronchi); the third one was related to regional air-volume change such as  $U/(M+L)|v$ ; the fourth one was related to  $AirT\%$ ; the fifth one was related to  $D_h^*$  in Trachea; the sixth one was related to  $WT^*$  of BronInt and  $D_h^*$  in sRUL; the seventh one was related to  $Cr$  and  $\Delta V_{air}^F$  in RML; the eighth one was related to  $Cr$  of BronInt. Using the principal components rather than a set of original variables is advantageous for the cluster analysis, because this not only avoids multiple use of the correlated variables, but it also increases the clustering stability by weighting the variance of the principal components.

### Clustering Analysis

We compared three different clustering methods, i.e, hierarchical, *K-means* and Gaussian finite mixture model-based (E19), in order to find the best clustering method with respect to internal properties of clusters (**Figure E2a**). First, we evaluated the internal properties of clusters using three quantities, namely, *connectivity*, *Silhouette width*, and *Dunn index* (E20). The quantities indicated the degree of connectedness of the clusters, the degree of confidence in particular clustering assignment, and the ratio of the smallest distance between observations not in the same cluster to the largest intra-cluster distance, respectively. Smaller *connectivity* and larger *Silhouette width* and *Dunn index* indicate the better quality of clustering. A package “clValid” in software *R* (E20) was used to perform this analysis. **Figure E2a** shows that model-based clustering is not a good choice in the current imaging data with respect of internal properties. Based upon *Dunn index*, the number of 4 or 5 are likely to be a potential candidate for the number of clusters when using either hierarchical or *K-means*.

We then performed a cluster stability analysis in four different cases: *K-means* with 4 clusters, *K-means* with 5 clusters, Hierarchical with 4 clusters, and Hierarchical with 5 clusters

(**Figure E2b**). For this analysis, we performed a non-parametric bootstrap analysis, based on resampling from the raw imaging data (E21). We performed cluster analysis on each of two hundred bootstrapped datasets, and the mean of Jaccard similarity coefficients, defined by the size of intersection divided by the size of the union between clusters, was calculated to compare the cluster patterns derived from the bootstrapped datasets with the original clusters. *K-means* clustering provided much more stable results than Hierarchical clustering (**Figure E2b**), where all clusters derived by *K-means* remained in the stable regions ( $> 0.75$ ) (E21). We further performed an analysis to find an optimal number of clusters. The only number of “4” in *K-means* was supported by five different statistical approaches (E22-26). A package “NbClust” in software *R* (E27) was used to perform all these methods. Lastly, we plotted clustering membership on two projected dimensional coordinates using multi-dimensional scaling technique (E28) to visualize the clustering results (**Figure E2c and d**). *K-means* clustering with each cluster demonstrated clear separation, whereas the Cluster 1 of Hierarchical clustering was scattered in other clusters.

### Association with “Simple” Clinical Clusters of Asthma

The imaging-based clusters were compared with “simple” clinical clusters derived by Moore et al. (E29) based upon three variables of onset of asthma, baseline and maximal FEV<sub>1</sub>. Our imaging clusters, compared with the earlier “simple” clinical clusters, demonstrated a moderate Spearman’s rank correlation of  $\rho = 0.319$  with  $P < 0.0001$  (**Figure E3**) and chi-square association of  $P < 0.0001$ . Specifically, 77% of Cluster 1 was categorized into simple Cluster 1 or 2 close to normal pre- and post- bronchodilator FEV<sub>1</sub>, whereas 73% of subjects in the imaging-based Clusters 4 were categorized into simple Cluster 4 or 5 which were characterized by the decrease of pre-bronchodilator FEV<sub>1</sub> (E29).

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## SUPPLEMENTARY TABLES

Table E1. Standardized loadings of eight principal components based upon correlation matrix.

The variables with the significant correlation in each component are indicated by an *asterisk*.

Full names of each variable were described in *Abbreviations used* of the main text. These

variables consist of 32 local ( $\theta$  in two regions,  $Cr$ ,  $WT$  and  $D_h^*$  in ten regions), 20 lobar ( $\Delta V_{air}^F$ ,

$AirT\%$ , Jacobian and ADI in five lobes) and 5 global (lung shape,  $U/(M+L)|v$ ,  $AirT\%$ , Jacobian

and ADI) variables. Among 57 variables, 4 variables including  $WT^*$  (sRUL),  $D_h^*$  (sLLL),  $AirT\%$  (Total) and Jacobian (Total) used to derive simple imaging-based clusters (**Figure 3**).

|                         |  | Principal components |     |     |     |     |     |     |     |
|-------------------------|--|----------------------|-----|-----|-----|-----|-----|-----|-----|
|                         |  | 1                    | 2   | 3   | 4   | 5   | 6   | 7   | 8   |
| Proportion of variation |  | 25%                  | 12% | 10% | 6%  | 5%  | 4%  | 3%  | 3%  |
| Cumulative variation    |  | 25%                  | 37% | 47% | 53% | 58% | 62% | 65% | 68% |

| Variable | Region      | Standardized loadings based upon correlation matrix |               |               |       |               |              |              |              |
|----------|-------------|-----------------------------------------------------|---------------|---------------|-------|---------------|--------------|--------------|--------------|
| $\theta$ | Trachea     | 0.02                                                | -0.07         | 0.42          | -0.09 | 0.34          | -0.09        | 0.25         | -0.27        |
|          | RMB         | -0.10                                               | -0.06         | -0.27         | -0.08 | -0.10         | -0.25        | -0.33        | -0.26        |
| $Cr$     | Trachea     | -0.27                                               | -0.07         | -0.44         | 0.14  | -0.41         | 0.13         | -0.01        | 0.15         |
|          | RMB         | -0.32                                               | -0.06         | <b>-0.51*</b> | 0.12  | -0.22         | 0.20         | -0.03        | 0.46         |
|          | BronInt     | -0.14                                               | -0.03         | -0.20         | -0.02 | 0.17          | 0.34         | 0.00         | <b>0.61*</b> |
|          | LMB         | -0.26                                               | -0.05         | -0.33         | -0.10 | -0.17         | 0.33         | 0.11         | 0.36         |
|          | TriLLB      | -0.15                                               | -0.14         | -0.29         | -0.11 | 0.10          | 0.18         | 0.25         | 0.06         |
|          | sRUL        | -0.12                                               | 0.08          | -0.18         | 0.01  | -0.09         | 0.02         | 0.20         | 0.09         |
|          | sRML        | -0.16                                               | -0.18         | -0.12         | -0.17 | -0.05         | -0.13        | <b>0.53*</b> | -0.12        |
|          | sRLL        | -0.10                                               | -0.01         | -0.13         | -0.11 | 0.11          | 0.07         | 0.47         | -0.04        |
|          | sLUL        | -0.21                                               | -0.19         | -0.22         | -0.18 | -0.14         | -0.23        | 0.38         | 0.06         |
|          | sLLL        | -0.15                                               | -0.19         | -0.16         | -0.17 | 0.03          | 0.13         | 0.32         | 0.00         |
| $WT^*$   | Trachea     | -0.15                                               | -0.33         | 0.44          | -0.05 | 0.21          | 0.17         | -0.04        | -0.18        |
|          | RMB         | -0.27                                               | -0.28         | 0.10          | -0.05 | -0.39         | 0.12         | 0.08         | -0.15        |
|          | BronInt     | -0.18                                               | -0.45         | 0.31          | -0.08 | 0.27          | <b>0.39*</b> | -0.13        | 0.01         |
|          | LMB         | -0.19                                               | -0.31         | 0.13          | 0.04  | -0.21         | 0.33         | -0.17        | -0.24        |
|          | TriLLB      | -0.26                                               | -0.52         | 0.42          | 0.00  | 0.33          | 0.21         | -0.13        | 0.00         |
|          | <b>sRUL</b> | -0.25                                               | -0.43         | 0.55          | 0.09  | 0.30          | 0.02         | -0.20        | 0.14         |
|          | sRML        | -0.38                                               | -0.51         | 0.19          | 0.04  | 0.27          | -0.04        | 0.17         | 0.07         |
|          | sRLL        | -0.41                                               | -0.55         | 0.29          | 0.10  | 0.40          | -0.07        | -0.04        | 0.08         |
|          | sLUL        | -0.32                                               | -0.56         | 0.34          | 0.17  | 0.15          | -0.15        | -0.03        | 0.23         |
|          | sLLL        | -0.31                                               | <b>-0.61*</b> | 0.32          | 0.00  | 0.44          | 0.03         | -0.04        | 0.09         |
| $D_h^*$  | Trachea     | -0.36                                               | -0.35         | 0.05          | 0.11  | <b>-0.55*</b> | 0.25         | -0.08        | -0.31        |
|          | RMB         | -0.57                                               | -0.38         | -0.05         | 0.24  | -0.41         | 0.28         | -0.07        | -0.09        |

|                           |              |               |               |               |               |       |               |              |       |
|---------------------------|--------------|---------------|---------------|---------------|---------------|-------|---------------|--------------|-------|
|                           | BronInt      | -0.52         | -0.38         | 0.09          | 0.17          | -0.21 | 0.36          | -0.05        | -0.04 |
|                           | LMB          | -0.46         | -0.42         | 0.11          | 0.07          | -0.44 | 0.30          | -0.02        | -0.26 |
|                           | TriLLB       | <b>-0.68*</b> | -0.45         | -0.10         | 0.19          | -0.14 | -0.12         | 0.04         | 0.02  |
|                           | sRUL         | -0.56         | -0.41         | 0.08          | 0.27          | -0.22 | <b>-0.39*</b> | -0.08        | 0.10  |
|                           | sRML         | -0.58         | -0.43         | -0.11         | 0.22          | -0.10 | -0.26         | 0.29         | 0.01  |
|                           | sRLL         | -0.64         | -0.44         | -0.13         | 0.21          | -0.08 | -0.31         | 0.01         | 0.03  |
|                           | sLUL         | -0.59         | -0.46         | 0.04          | 0.26          | -0.18 | -0.37         | 0.04         | 0.14  |
|                           | <b>sLLL</b>  | -0.63         | <b>-0.52*</b> | -0.14         | 0.19          | -0.08 | -0.27         | 0.04         | 0.01  |
| Shape at TLC              |              | -0.20         | 0.10          | -0.51         | -0.04         | -0.12 | -0.26         | -0.31        | 0.00  |
| U/(M+L) v                 |              | 0.12          | 0.42          | <b>0.74*</b>  | 0.26          | -0.33 | -0.07         | 0.00         | 0.15  |
| $\Delta V_{\text{air}}^F$ | LUL          | 0.00          | 0.46          | 0.62          | 0.30          | -0.31 | -0.06         | 0.14         | 0.11  |
|                           | LLL          | -0.02         | -0.44         | <b>-0.64*</b> | -0.24         | 0.24  | 0.13          | -0.11        | -0.03 |
|                           | RUL          | 0.14          | 0.34          | 0.71          | 0.19          | -0.29 | -0.06         | -0.09        | 0.19  |
|                           | RML          | -0.17         | 0.14          | 0.16          | 0.10          | 0.06  | 0.18          | <b>0.54*</b> | -0.21 |
|                           | RLL          | 0.00          | -0.33         | -0.62         | -0.24         | 0.25  | -0.14         | -0.23        | -0.12 |
| AirT%                     | <b>Total</b> | 0.60          | -0.44         | 0.22          | <b>-0.56*</b> | -0.26 | -0.06         | 0.02         | 0.09  |
|                           | LUL          | 0.59          | -0.47         | 0.14          | -0.51         | -0.21 | -0.04         | 0.04         | 0.05  |
|                           | LLL          | 0.52          | -0.33         | 0.25          | -0.50         | -0.27 | -0.10         | -0.04        | 0.20  |
|                           | RUL          | 0.57          | -0.47         | 0.10          | -0.51         | -0.23 | -0.05         | 0.03         | 0.06  |
|                           | RML          | 0.57          | -0.40         | 0.24          | -0.53         | -0.19 | -0.06         | 0.04         | -0.02 |
|                           | RLL          | 0.53          | -0.33         | 0.27          | -0.52         | -0.29 | -0.09         | -0.04        | 0.18  |
| Jacobian                  | <b>Total</b> | <b>-0.87*</b> | 0.29          | 0.01          | -0.31         | 0.05  | 0.03          | -0.03        | 0.04  |
|                           | LUL          | -0.83         | 0.35          | 0.10          | -0.24         | 0.02  | 0.03          | 0.00         | 0.07  |
|                           | LLL          | <b>-0.85*</b> | 0.20          | -0.13         | -0.33         | 0.05  | 0.03          | -0.05        | -0.01 |
|                           | RUL          | -0.81         | 0.34          | 0.14          | -0.27         | 0.05  | 0.04          | -0.01        | 0.09  |
|                           | RML          | -0.81         | 0.33          | 0.09          | -0.26         | 0.03  | 0.04          | 0.07         | 0.05  |
|                           | <b>RLL</b>   | <b>-0.86*</b> | 0.22          | -0.10         | -0.35         | 0.06  | 0.01          | -0.06        | -0.03 |
| ADI                       | Total        | <b>-0.85*</b> | 0.28          | 0.16          | -0.33         | -0.04 | -0.06         | -0.06        | -0.03 |
|                           | LUL          | <b>-0.78*</b> | 0.33          | 0.27          | -0.22         | -0.04 | -0.05         | 0.02         | 0.05  |
|                           | LLL          | -0.78         | 0.25          | 0.01          | -0.33         | -0.07 | -0.08         | -0.05        | -0.08 |
|                           | RUL          | -0.70         | 0.27          | 0.41          | -0.22         | -0.01 | -0.03         | -0.01        | 0.07  |
|                           | RML          | -0.65         | 0.11          | 0.17          | -0.37         | -0.03 | -0.07         | -0.08        | -0.05 |
|                           | <b>RLL</b>   | <b>-0.79*</b> | 0.21          | 0.04          | -0.34         | -0.06 | -0.05         | -0.13        | -0.10 |

**SUPPLEMENTARY FIGURES CAPTIONS**

Figure E1. A scree plot: Eigenvalues (magnitudes of variances) according to the number of principal components for determining the optimal number of principal components

Figure E2. Clustering analysis, a: Internal property in different clustering methods; b: Clustering stability analysis between *K*-means and Hierarchical clustering with 4 or 5 number of clusters; c: Clustering membership of *K*-means clustering on 2-D projected coordinates; d: Clustering membership of Hierarchical clustering on 2-D projected coordinates

Figure E3. Relationship of imaging-based clusters vs. simple clinical clusters by Moore et al. (2010, *Am J Respir Crit Care Med*); Spearman's correlation  $r = 0.319$  ( $P < 0.0001$ ).