

Images in Allergy and Immunology

Susan Foley, MD, and Qutayba Hamid, MD, PhD, Editors

Severe asthma

Editor's note: This feature, Images in allergy and immunology, is designed to highlight current concepts of the immunopathology of allergic diseases and other common immunologically mediated diseases. The presentation will appear as sets of images that involve cross-pathology, histopathology, and molecular pathology and will cover a range of topics of interest to allergists and immunologists.

Subjects with severe asthma comprise 10% or less of the asthmatic population but are responsible for a disproportionately high fraction of the health care budget. They appear to have different airway behavior than subjects with mild and moderate asthma, as evidenced by persistently low expiratory flow rates and failure to respond adequately to treatment in terms of pulmonary function.¹ In addition, they often remain clinically symptomatic despite maximal medical therapy, and their disease remains difficult to control.

Radiologically, bronchial wall thickening, expiratory air trapping, and bronchial lumen narrowing are the most consistent high-resolution computed tomographic features reported in severe asthma (Fig 1).² However, considerable overlap exists between the spectrum of clinical and functional features of patients with bronchiolitis obliterans and those with severe asthma, especially in asthmatic subjects whose airway obstruction is incompletely reversible. In some cases the 2 diseases might be indistinguishable radiologically.

Wenzel et al³ suggested that the phenotype of severe asthma comprises at least 2 different and distinct pathologic subtypes on the basis of the presence or absence of eosinophils in bronchoalveolar lavage (BAL) fluid.

From Meakins-Christie Laboratories, McGill University, Montreal, Quebec, Canada.

Received for publication November 14, 2005; revised December 6, 2005; accepted for publication December 6, 2005.

Reprint requests: Qutayba Hamid, MD, PhD, Meakins-Christie Laboratories, 3626 St Urbain St, Montreal H2X2P2, Quebec, Canada. E-mail: qutayba.hamid@mcgill.ca.

J Allergy Clin Immunol ■■■■■■■■

0091-6749/\$32.00

© 2006 American Academy of Allergy, Asthma and Immunology

doi:10.1016/j.jaci.2005.12.1302

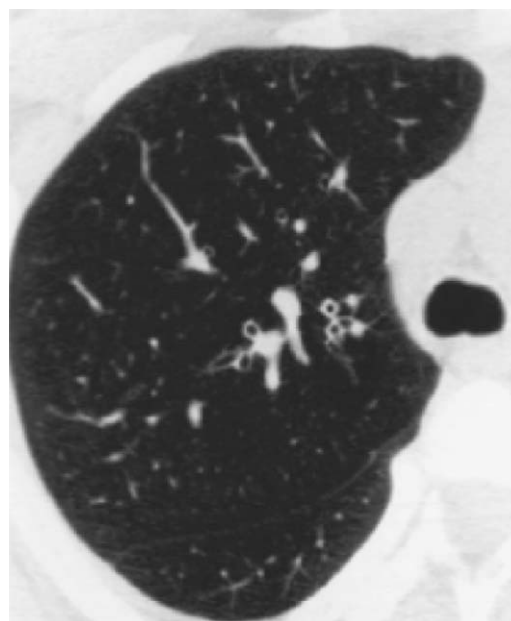


FIG 1. Inspiratory high-resolution computed tomography through the right upper lobe of a subject with severe asthma, demonstrating bronchial wall thickening. Reprinted from Jensen et al² with permission from The Royal College of Radiologists.

The subtype with eosinophil predominance is associated with increased subbasement membrane thickening compared with the subtype with relatively few eosinophils and compared with subjects with mild asthma and control subjects. Clinically and physiologically, however, the 2 subtypes of severe asthma behave similarly. Intense eosinophilic infiltrates can also be observed distally in the small airways in patients with chronic severe asthma (Fig 2).

Increased numbers of neutrophils have also been reported in BAL fluid and endobronchial and trans-bronchial biopsy specimens of subjects with refractory asthma^{3,4} when compared with those with milder disease, indicating that there is ongoing inflammation in the airways of patients with severe asthma with poor airway function, despite very high doses of oral and inhaled steroids. Neutrophil numbers are similar in the 2 subgroups of severe asthma.

Remodeling in asthma comprises a number of structural changes, including epithelial detachment, subepithelial fibrosis, and smooth muscle hyperplasia and hypertrophy, all of which might contribute to irreversibility of airflow obstruction with disease progression. Autopsy studies have shown that the entire length of the airway is in fact involved in severe fatal asthma (Fig 3).⁵⁻⁸ Alteration in smooth muscle has been reported as the main structural change that distinguishes severe from moderate asthma.⁹ Pepe et al⁹ demonstrated that airway smooth muscle area was greater in subjects with severe asthma when compared with that seen in those with moderate disease and that

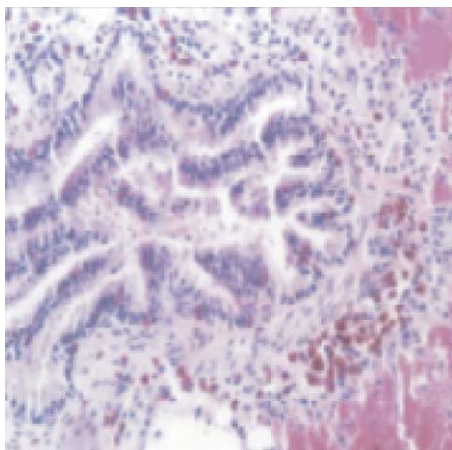


FIG 2. This cross-section of a small airway from a patient with chronic severe asthma shows eosinophilic infiltrate in the submucosa and in the area of the smooth muscle. Reprinted from Hamid Q. Gross pathology and histopathology of asthma. *J Allergy Clin Immunol* 2003;111:431-2, with permission from the American Academy of Allergy, Asthma, and Immunology.

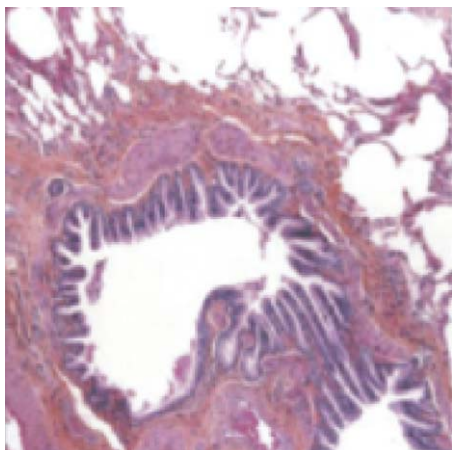


FIG 3. Cross-section of a small airway from a patient who died of complications of severe chronic asthma. This section shows extensive small airway remodeling with increased smooth muscle mass and collagen deposition. Reprinted from Hamid Q. Gross pathology and histopathology of asthma. *J Allergy Clin Immunol* 2003;111:431-2, with permission from the American Academy of Allergy, Asthma, and Immunology.

the airway smooth muscle was closer to the epithelium in the more severely diseased group (Fig 4, A). The greater muscle mass also had a more activated phenotype in patients with severe asthma, as evidenced by the increased expression of IL-8 and eotaxin (Fig 4, B). These cytokines are likely to be pivotal in the ongoing inflammatory cycle that characterizes the airways of those with severe refractory asthma.

A number of profibrotic cytokines are increased in severe asthma and are associated with the remodeling that occurs with disease progression. Minshall et al¹⁰ reported increased deposition of extracellular matrix proteins, particularly collagens I and III (Fig 5, A and

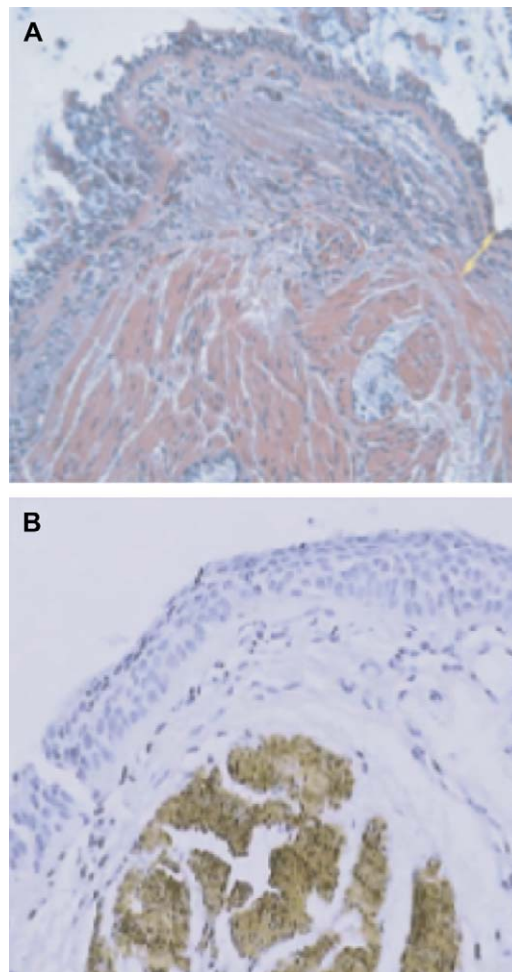


FIG 4. A, Hematoxylin and eosin staining of a bronchial biopsy specimen from a subject with severe asthma showing increased smooth muscle mass with decreased distance between the smooth muscle and epithelial layers. B, Representative example of immunostaining for eotaxin in the airway smooth muscle of a subject with severe asthma. Reprinted with permission from Pepe et al.⁹

B), as well as increased IL-11 expression (Fig 5, C), in subjects with severe asthma compared with that seen in subjects with mild asthma and control subjects. IL-11, a pleiotropic cytokine with profibrogenic and potent immunomodulatory properties, was found to be increased, particularly within the bronchial mucosa and airway epithelial cells of subjects with severe disease. Within the subepithelial cell layer, IL-11 mRNA colocalized to major basic protein-positive eosinophils (Fig 5, D). There was an inverse correlation between the number of cells expressing IL-11 mRNA and the FEV₁ of the subjects, suggesting that the release of IL-11 from the epithelium and subepithelial tissues might contribute both to the development of airway fibrosis and the subsequent decrease in lung function. The expression of IL-17 is also upregulated in severe asthma in a pattern similar to that seen with IL-11, and treatment with oral corticosteroids

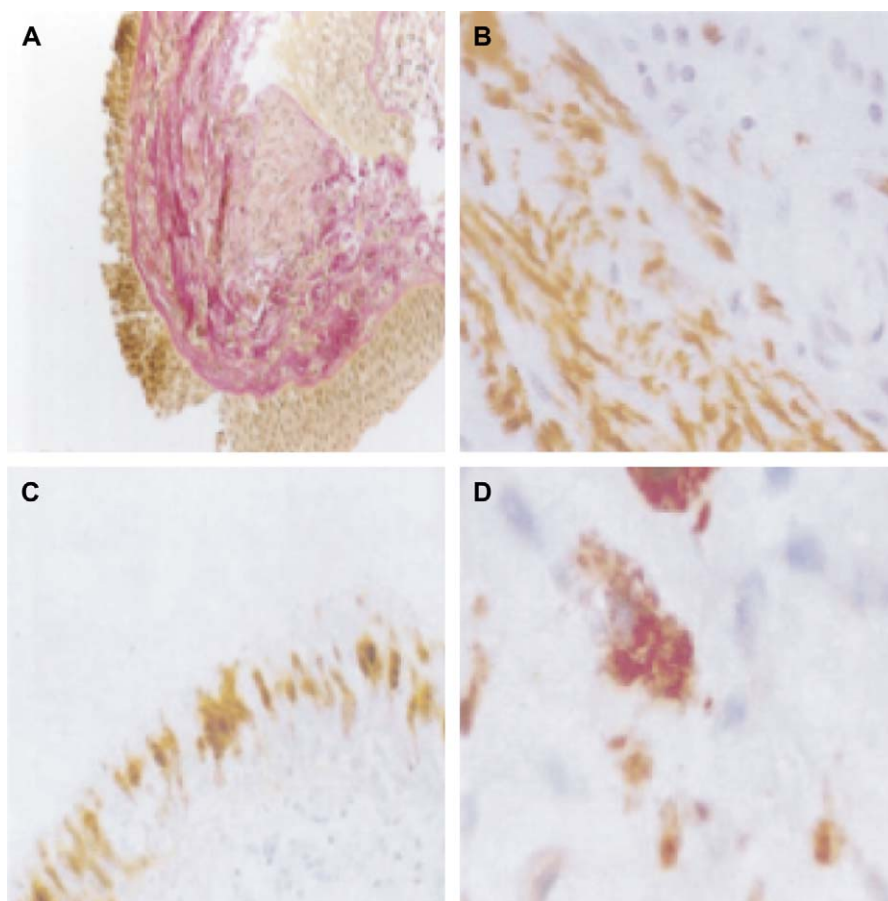


FIG 5. **A**, Van Gieson staining showing increased total collagen (pink staining) in a subject with severe asthma. **B**, Collagen type III immunoreactivity, as determined by using the avidin-biotin peroxidase method, in a subject with severe asthma is shown as brown staining. **C**, Immunostaining for IL-11 in the epithelium of an individual with severe asthma. **D**, Colocalization of IL-11 immunoreactivity and eosinophil-specific marker (major basic protein) in a subject with severe asthma. IL-11 immunoreactivity is shown as brown coloration that localizes to major basic protein-positive cells (red staining). Reprinted with permission from Minshall et al.¹⁰

significantly reduces the expression of both these cytokines in the airways of subjects with severe asthma.¹¹ Balzar et al¹² recently identified TGF- β 2 as the predominant TGF- β isoform in severe asthma. It is expressed mainly by eosinophils and is associated with increased profibrotic responses.

Steroid-resistant asthma refers to a subset of asthmatic patients whose symptoms do not respond even to systemic glucocorticoid therapy. Leung et al¹³ demonstrated that steroid-resistant asthma is associated with a dysregulation of the expression of the genes encoding for T_H1/T_H2 cytokines in airway cells. Their study gave further strength to the concept that the combination of IL-2 and IL-4 reduces glucocorticoid receptor (GR) binding affinity and T-cell responsiveness to glucocorticoids.¹⁴

Glucocorticoids exert their effect through ligation with the functional isoform GR α . Patients with steroid-resistant asthma have significantly higher numbers of GR β -immunoreactive cells in peripheral blood and BAL fluid compared with steroid-sensitive

patients or control subjects, suggesting that GR β (a dominant negative inhibitor of steroid action) might be a marker of steroid insensitivity.¹⁵ Indeed, there is significantly higher airway expression of GR β in both the large and small airways in patients with severe fatal asthma compared with that seen in patients with mild asthma and control subjects, alluding to a close association between GR β and steroid insensitivity in severe asthma. We have recently shown (submitted manuscript) that the GR β isoform is overexpressed in the airway epithelium and submucosal inflammatory cells of patients with severe compared with moderate asthma, resulting in a lower GR α /GR β ratio. This might explain the difficulty in achieving adequate disease control with inhaled corticosteroids in the more severely diseased group.

REFERENCES

1. Bumbacea D, Campbell D, Nguyen L, Carr D, Barnes PJ, Robinson D, et al. Parameters associated with persistent airflow obstruction in chronic severe asthma. *Eur Respir J* 2004;27:1-2.

2. Jensen SP, Lynch A, Brown KK, Wenzel SE, Newell JD. High-resolution CT features of severe asthma and bronchiolitis obliterans. *Clin Radiol* 2002;57:1078-85.
3. Wenzel SE, Schwartz LB, Langmack EL, Halliday JL, Trudeau JB, Gibbs RL, et al. Evidence that severe asthma can be divided pathologically into two inflammatory subtypes with distinct physiologic and clinical characteristics. *Am J Respir Crit Care Med* 1999;160:1001-8.
4. Wenzel SE, Szefer SJ, Leung DYM, Sloan SI, Rex MD, Martin RJ. Bronchoscopic evaluation of severe asthma. Persistent inflammation associated with high dose glucocorticoids. *Am J Respir Crit Care Med* 1997;156:737-43.
5. Carroll N, Cooke C, James A. The distribution of eosinophils and lymphocytes in the large and small airways of asthmatics. *Eur Respir J* 1997;10:292-300.
6. Faul JL, Tormey VJ, Leonard C, Burke CM, Farmer J, Horne SJ, et al. Lung immunopathology in cases of sudden asthma death. *Eur Respir J* 1997;10:301-7.
7. Kuwano K, Bosken CH, Pare PD, Bai TR, Wiggs BR, Hogg JC. Small airways dimensions in asthma and in chronic obstructive pulmonary disease. *Am Rev Respir Dis* 1993;148:1220-5.
8. Woolcock AJ. Effects of drugs on small airways. *Am J Respir Crit Care Med* 1998;157(suppl):203-7.
9. Pepe C, Foley S, Shannon J, Lemiere C, Olivenstein R, Ernst P, et al. Differences in airway remodeling between subjects with severe and moderate asthma. *J Allergy Clin Immunol* 2005;116:544-9.
10. Minshall E, Chakir J, Laviolette M, Molet S, Zhu Z, Olivenstein R, et al. IL-11 expression is increased in severe asthma: association with epithelial cells and eosinophils. *J Allergy Clin Immunol* 2000;105:232-8.
11. Chakir J, Shannon J, Molet S, Fukakusa M, Elias J, Laviolette M, et al. Airway remodeling-associated mediators in moderate to severe asthma: Effect of steroids on TGF β , IL-11, IL-17 and type I and type III collagen expression. *J Allergy Clin Immunol* 2003;111:1293-8.
12. Balzar S, Chu HW, Silkoff P, Cundall M, Trudeau JB, Strand M, et al. Increased TGF- β 2 in severe asthma with eosinophilia. *J Allergy Clin Immunol* 2005;115:110-7.
13. Leung DYM, Martin RJ, Szefer SJ, Sher ER, Ying S, Kay AB, et al. Dysregulation of interleukin 4, interleukin 5 and interferon γ expression in steroid-resistant asthma. *J Exp Med* 1995;181:33-40.
14. Kam JC, Szefer SJ, Surs W, Sher ER, Leung DYM. Combination IL-2 and IL-4 reduces glucocorticoid receptor-binding affinity and T cell response to glucocorticoids. *J Immunol* 1993;151:3460-6.
15. Leung DYM, Hamid Q, Vottero A, Szefer SJ, Surs W, Minshall E, et al. Association of glucocorticoid insensitivity with increased expression of glucocorticoid receptor β . *J Exp Med* 1997;186:1567-74.