

Evidence of a genetic contribution to lung function decline in asthma

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There has been great progress in identifying new asthma susceptibility genes. In asthmatic subjects there is variable airway remodeling that includes features such as smooth muscle hypertrophy/hyperplasia, basement membrane thickening, and increased extracellular matrix deposition. Does airway remodeling have a genetic contribution in asthma? Data from different murine strains suggest there is a genetic contribution to the development and progression of airway remodeling. In human subjects it is important to consider what surrogate markers of remodeling have been used in genetic studies. Baseline FEV₁ and airway hyperresponsiveness are determined by a complex interplay of factors, including nonremodeling mechanisms; however, we consider a decline in FEV₁ as a robust marker of remodeling. To date, single nucleotide polymorphisms spanning *ADAM33*, *ESR1*, *PLAUR*, and *VEGF* have been associated with an excess decline in lung function in asthmatic subjects carrying the rare alleles (FEV₁, –13.0 to 55.2 mL/y excess). Interestingly these genes have overlapping functions in proteolytic pathways in the airways. There is accumulating evidence that genetic factors are important in the development of airway remodeling in asthmatic subjects, and further longitudinal studies with additional remodeling phenotypes and genome-wide association studies will identify novel susceptibility genes, leading to new approaches to target remodeling in asthmatic subjects. (J Allergy Clin Immunol 2011;■■■:■■■-■■■.)

Key words: Asthma, airway remodeling, genetic susceptibility, lung function decline

Asthma is a chronic inflammatory disease that is characterized by respiratory symptoms, variable airway obstruction, and airways hyperresponsiveness (AHR). Asthma involves the interaction of genetic and environmental components leading to disease, with heritability estimates varying between 36% to 79%.¹ Recent

Abbreviations used

ADAM33:	A disintegrin and metalloproteinase domain 33
AGER:	Advanced glycosylation end-product receptor
AHR:	Airways hyperresponsiveness
ASM:	Airway smooth muscle
ERK:	Extracellular signal-regulated kinase
ESR1:	Estrogen receptor 1
FVC:	Forced vital capacity
GWA:	Genome-wide association
HHIP:	Hedgehog interacting protein
LD:	Linkage disequilibrium
OVA:	Ovalbumin
PAI:	Plasminogen activator inhibitor
PAS:	Plasminogen activation system
PLAUR:	Urokinase plasminogen activator receptor
SNP:	Single nucleotide polymorphism
VEGF:	Vascular endothelial growth factor

years have shown rapid progress in mapping novel asthma loci by using genome-wide association (GWA) approaches, such as chromosome 17q21 (including *ORMDL3*), *IL33*, *IL1RL1/IL18R1*, *HLA-DQ*, *IL2RB*, *SMAD3*, *PDE4D*, *RAD50*, and *DENND1B*.² These findings expand on previously identified genes using positional cloning, such as a disintegrin and metalloproteinase domain 33 (*ADAM33*), *NPSR1*, *HLA-G*, protocadherin-1 (*PCDH1*), and urokinase plasminogen activator receptor (*PLAUR*).² Thus far, it remains largely unknown how these genes predispose subjects to specific clinical phenotypes that contribute to the expression and progression of asthma. The observation that many novel asthma genes are specifically expressed in airway structural cells suggests an important role for airway cells in asthma development.³ In this review we consider airway remodeling as a specific disease process leading to subphenotypes of asthma, such as accelerated lung function decline over time. We will discuss the current evidence supporting a role for genetic factors in predisposing subjects to airway remodeling.

AIRWAY REMODELING IN ASTHMA

Structural alterations termed “airway remodeling” occur in asthmatic subjects, and the extent of remodeling is variable. Airway remodeling is characterized by smooth muscle hypertrophy/hyperplasia, subepithelial fibrosis, basement membrane thickening, increased extracellular matrix deposition, and angiogenesis, and more recently, alterations in epithelial functions have been identified, including metaplasia and goblet cell hyperplasia.⁴ These structural changes have been observed in both children and adults with asthma, suggesting they are an early feature of the disease.⁵ The consequences of airway remodeling include

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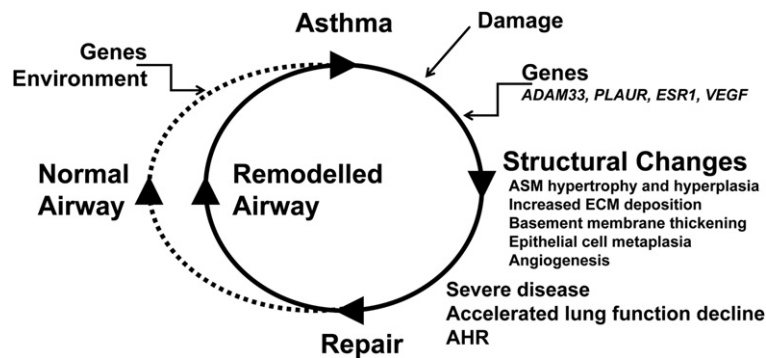


FIG 1. Possible mechanisms underlying airway remodeling in asthmatic subjects. A combination of genetic and environmental factors leads to the development of asthma. In those subjects with genetic predisposition to airway remodeling, damage to the airways leads to structural changes, which manifest in clinical outcomes, including accelerated decline in lung function, severe disease, and AHR. Airway-remodeling genes include those genes showing association with lung function decline in asthmatic subjects. *ECM*, Extracellular matrix.

increased AHR, fixed airway obstruction, and an accelerated decline in lung function. It remains to be determined how airway remodeling in asthmatic children affects lung growth and whether reduced lung growth is a marker of remodeling as well. Importantly, the degree of airway remodeling has been associated with the severity of asthma; for example, airway smooth muscle (ASM) mass is increased in subjects with severe versus moderate asthma.⁶ The mechanisms underlying the remodeling process remain to be defined. It is thought that this process is initiated by epithelial barrier damage as a result of inflammation caused by allergic or noxious stimuli or mechanical stress leading to aberrant repair in genetically susceptible subjects (Fig 1). These repair responses are associated with a series of alterations in resident cells of the airways, such as epithelial cells, fibroblasts, and ASM cells. For example, epithelial cells differentiate into mucus-producing goblet cells; fibroblasts develop an altered secretory profile, including production of collagens III and V; and fibronectin and smooth muscle develop a proliferative phenotype. It has also been suggested that epithelial mesenchymal transition is a feature of these processes.⁷ Therefore we consider airway remodeling to be a key feature of asthma that might distinguish how asthma presents in different subjects, potentially involving a distinct set of genes and pathways important in those genetically predisposed subjects.

EVIDENCE FOR A GENETIC CONTRIBUTION TO AIRWAY REMODELING? MICE AND MEN

A genetic contribution to the development of structural remodeling of the airways in allergic airway disease has been exquisitely demonstrated by using 4 different strains of mice.⁸ Mice have an eosinophil-driven inflammation of the airways after immunization with ovalbumin (OVA)/alum (sensitization) followed by intranasal administration of OVA (challenge). However, when the murine strains C57BL/6, BALB/C, C3H/HeJ, and A/J were compared, both overlapping and distinct features of airway remodeling were induced. Of particular relevance, collagen deposition, a feature of remodeling, was more prominent in A/J mice compared with that seen in the other strains of mice over a 12-week chronic OVA exposure period. Interestingly, bronchial airway wall thickening was also most prominent in A/J mice, with

some thickening seen in BALB/c mice but a total absence in the C57BL/6 and C3H/HeJ strains over the same period.⁸ AHR (methacholine) was present in all strains; however, it was significantly increased in the A/J mice that had features of airway remodeling. More recently, a similar study has confirmed these strain-dependent differences; for example, there was significantly greater collagen and elastic fiber content in lung parenchymal samples from BALB/c and A/J mice compared with C57BL/10 mice at baseline and during OVA sensitization/challenge.⁹

These data clearly demonstrate that the genetic background is important for the initiation and extent of airway remodeling in allergic airway disease, albeit in a murine model. They also highlight the complex interplay between different outcome measures that are genetically driven. For example, the A/J mice showed an increase in both remodeling and AHR measures, and in contrast, the BALB/c showed increased remodeling without an increase in AHR.

In human subjects, because of the limitations of invasive measures, there are no hereditary estimates for basement membrane thickening in asthma, although there is extensive evidence that surrogate markers of airway remodeling, namely baseline FEV₁ and AHR, have a clear genetic component. Hereditary estimates for baseline FEV₁ have been reported to be as high as 77%.¹⁰ Similarly, in asthmatic twin studies AHR (methacholine) showed a greater correlation in monozygotic twins, with an intra-pair correlation coefficient of 0.67 compared with 0.34 for dizygotic twins.¹¹ In asthmatic subjects there is evidence that differences in cell functions that are potentially genetically driven underlie airway-remodeling mechanisms. A recent study further confirmed dysregulated repair in epithelial cells isolated from asthmatic subjects, and expression array and functional analyses identified defective fibronectin expression as at least in part underlying this mechanism.¹² This provides further support that genetic mechanisms driving altered gene expression underlie the remodeling process.

PHENOTYPIC MARKERS OF AIRWAY REMODELING FOR GENETIC STUDIES

The multicellular nature and interplay of different mechanisms make airway remodeling a complex phenotype for genetic

TABLE I. Genes associated with accelerated lung function decline in asthmatic subjects

Gene	Key SNPs	Functional significance	Association	Reference
<i>ADAM33</i> *	rs258557G/C(S_2)	Gly/Gly in transmembrane domain	Excess FEV ₁ decline	Jongepier et al ²¹
	rs2280091A/G(T_1)	Met/Thr in cytoplasmic domain	Rare alleles: -46.9 to 55.2 mL/y	
	rs2280090G/A(T_2)	Pro/Ser in cytoplasmic domain		
<i>ESR1</i> *	rs4870056A/G (IVS1-1505)	Intron 1, regulatory?	Excess FEV ₁ decline	Dijkstra et al ²²
	rs9322331T/C (IVS1-1415)	Intron 1, regulatory?	Rare alleles: -13.0 to 16.1 mL/y	
	rs2234693 C/T (IVS1-397)	Intron 1, regulatory?	Female subjects more significant	
	rs9340799G/A (IVS1-351)	Intron 1, regulatory?	Haplotype: -25.8 to 29.2 mL/y	
<i>PLAUR</i> *	rs2077647T/C (IVS1+30)	Ser/Ser in exon 1		Barton et al ²³
	rs2356338G/T	-649 promoter, PLAUR levels	Excess FEV ₁ decline	
	rs4802189C/A	3' Region, function?	Rare alleles: -14.6 to 17.4 mL/y	
	rs4803648T/A	3' Region, function?		
<i>VEGF</i> †	rs4711750A/T	3' Region, function?	Excess FEV ₁ /FVC decline	Sharma et al ²⁴
<i>ARG1</i> *	rs2781667C/T	Intron 1, regulatory?	Excess FEV ₁ decline in the presence of corticosteroids	Vonk et al ²⁶

*The Dutch sample represents 200 adult asthmatic subjects followed for a mean of approximately 22 years with a median number of measurements of approximately 29 (see main text). Analyses in the Dutch population used a linear mixed effects models, and the excess decline values are shown in the table.

†Childhood Asthma Management Program subjects were followed for 4 years. Note that the decline in lung function in the general population is approximately 18.7 and 12.7 mL/y in female and male subjects, respectively.²⁵ *ARG1* is included for completeness, although this gene might be a modifier gene influencing corticosteroid effects on FEV₁ decline. No evidence exists that *ARG1* is a susceptibility gene for airway remodeling.²⁶

association studies. Studies assessing the genetic association of candidate genes with airway remodeling are very limited,¹³ and GWA studies of markers of airway remodeling have not been published to date. This has led to the use of surrogate markers that reflect ongoing pathology in the airways, such as baseline FEV₁, AHR, and decline in lung function.

The correlation of baseline FEV₁ with markers of airway remodeling remains controversial. For example, it has been shown that ASM mass negatively correlates with baseline FEV₁ in subjects with moderate and severe asthma.⁶ Moreover, baseline FEV₁ negatively correlates with subepithelial layer thickening¹⁴; however, there was no correlation between the ratio of intermediary bronchial wall thickness to outer diameter and FEV₁.¹⁵ Thus cross-sectional FEV₁ measurements have significant limitations as a marker of airway remodeling because they reflect a single time-point measurement of airway diameter that is determined by edema, mucus, thickening of the airway wall, and ASM contraction.

Similarly, AHR has been postulated to be a marker of ongoing remodeling processes in the airways; however, this correlation is complex. In a recent study of 50 subjects with severe asthma, there was a significant negative correlation between AHR (histamine) and ASM thickness; however, there was no correlation for AHR (methacholine) or FEV₁ with ASM thickness.¹⁶ We will therefore not consider cross-sectional studies of FEV₁ or AHR in this review.

The importance of FEV₁ decline in asthmatic subjects has been exemplified in seminal articles demonstrating an accelerated decline in asthmatic subjects. For example, FEV₁ was followed over a 15-year period in 1,095 asthmatic subjects from a total of 16,411 subjects. In male subjects there was an excess decline of -16 mL/y in asthmatic subjects versus control subjects.¹⁷ An excess decline in lung function in asthmatic subjects was also observed in the Busselton Health Study.¹⁸ A subsequent study showed that excess FEV₁ decline might be ameliorated by the use of corticosteroids.¹⁹ Finally, a subset of asthmatic subjects (ie, 15%) might have irreversible airflow obstruction. In this 26-year follow-up study, the development of irreversible airway obstruction was associated with low baseline FEV₁, less reversibility, and less severe AHR but not smoking.²⁰

GENETIC DETERMINANTS OF LUNG FUNCTION DECLINE IN ASTHMATIC SUBJECTS

Although we consider lung function decline (in milliliters per year) to be the most robust noninvasive indicator of airway remodeling in asthmatic subjects for genetic studies, very few studies have the longitudinal data in asthmatic subjects required for these analyses. The literature is therefore limited to candidate gene studies completed by a few research groups (Table I).²¹⁻²⁶ Our studies have used a unique cohort of 200 adult Dutch asthmatic probands identified in 1962 to 1975 and followed up to 30 years. All had AHR at baseline, and 81.9% were atopic. Associations between FEV₁ decline and single nucleotide polymorphisms (SNPs) spanning *ADAM33*,²¹ estrogen receptor 1 (*ESR1*),²² and *PLAUR*²³ have been reported by using this population. Similarly, in a study by Sharma et al,²⁴ longitudinal post-bronchodilator FEV₁/forced vital capacity (FVC) data spanning approximately 4 years identified an association with vascular endothelial growth factor (*VEGF*) polymorphisms.²⁴ This study was composed of 493 children with mild-to-moderate asthma with AHR.

ADAM33

By using a positional cloning approach in both asthma-enriched families and a case-control design, *ADAM33* was identified as an asthma susceptibility gene and a determinant of AHR and total IgE levels.²⁷ This association has been extensively replicated in additional populations with varying magnitudes of effect.² *ADAM33* is a complex protein and is a member of the zinc-dependent metalloproteinase superfamily ADAM, members of which regulate their own function and a wide range of other proteins through proteolytic cleavage.²⁸ *ADAM33* is a multidomain protein, including prodomain, catalytic, metalloprotease, disintegrin (binds integrins), cysteine-rich/epidermal growth factor (cell-cell contact), transmembrane, and cytoplasmic domains and importantly exists in multiple splice forms containing various combinations of these domains. *ADAM33* is expressed predominantly in airway structural cells, including airway epithelium,

ASM, myofibroblasts, and fibroblasts, and is thought to have a wide spectrum of functions, including protease-dependent and independent mechanisms. ADAMs cleave proteins from the cell surface and might facilitate release of cytokines and growth factors; similarly, ADAMs are thought to regulate cell-cell and cell-extracellular matrix interactions. Soluble ADAM33 has been implicated in angiogenesis.²⁹ Therefore on the basis of the expression profile and predicted function, it was postulated that ADAM33 might be involved in airway remodeling in asthmatic subjects.

As shown in Table I, 3 SNPs demonstrated an association with excess FEV₁ decline in asthmatic subjects, rs528557G/C(S₂), rs2280091A/G(T₁), and rs2280090G/A(T₂), which supports the hypothesis that genetic alterations in this protease predispose asthmatic subjects to remodeling.²¹ The rs528557G/C(S₂) and rs2280091A/G(T₁) SNPs are in high linkage disequilibrium (LD) and potentially reflect the same causative mechanism, although the functional basis of these transmembrane and cytoplasmic domain polymorphisms remains to be determined. It is important to note that ADAM33 SNPs have also shown association with excess FEV₁ decline in a general population sample (n = 1390) followed for 25 years.²⁵ The main SNPs identified included rs612709C/T(Q₁) (cysteine-rich domain), rs3918336G/A(S₁) (transmembrane domain), and rs528557G/C(S₂), with excess decline of -3.6 to 9.6 mL/y.²⁵ rs3918336G/A(S₁) (transmembrane domain) and rs528557G/C(S₂) were associated with chronic obstructive pulmonary disease in this population, reflecting the potential role of ADAM33 in airway remodeling in multiple respiratory diseases.

ESR1

It is well documented that asthma in male and female subjects presents differently at different times of life (ie, the prevalence of asthma is higher in male children); however, by adulthood, female subjects more frequently have asthma and more commonly have severe asthma. These observations led to the hypothesis that the mechanism underlying this increased susceptibility to severe asthma in adult female subjects is hormone driven and that *ESR1* polymorphisms might be of significance.

As shown in Table I, 5 SNPs within *ESR1* have been shown to be risk factors for excess decline in FEV₁ in asthmatic subjects (range, -13.0 to 16.1 mL/y), which was driven by the presence of haplotypes of these SNPs in female subjects (eg, rs2077647TT/rs2234693TT/rs9340799AA carriers have an excess decline in FEV₁ of -40.1 mL/y [vs homozygous reference]).²² These SNPs are intronic in *ESR1* and in high LD with the functional significance of the polymorphisms remaining to be resolved. Multiple *ESR1* SNPs, including rs9322331T/C (IVS1-1415), are also genetic determinants of active plasmin concentrations in women undergoing hormone replacement.³⁰ This links *ESR1* polymorphisms with the plasminogen activation system (PAS).

Plasmin has many downstream proteolytic effects, such as extracellular matrix degradation and matrix metalloproteinase activation, which might influence remodeling of the airways. Similarly, the plasmin pathway has been implicated in angiogenesis. We have identified the pivotal receptor in this pathway involved in the generation of plasmin (ie, *PLAUR*) as containing genetic determinants of decline in FEV₁ in asthmatic subjects (see below).²³ Both *ESR1* and *ESR2* are known to be expressed in the

lung. Importantly, structural cells, including lung myofibroblasts, express *ESR1*, and activation of these cells with estrogen leads to phosphorylation of Raf1 and extracellular signal-regulated kinase (ERK) 1/2 and a decrease in proliferation.³¹

PLAUR

We identified an association between multiple SNPs spanning the *PLAUR* gene and asthma susceptibility, decline in FEV₁, and *PLAUR* serum levels by using 587 asthma-enriched families.²³ *PLAUR* is the pivotal GPI-linked serine protease receptor in the PAS. The interaction between *PLAU*-*PLAUR* is critical for the conversion of plasminogen to plasmin (through activation of the protease activity of *PLAU*) and is regulated by a series of proteins, including plasminogen activator inhibitor (PAI) 1, PAI-2, and PAI-3. Plasmin has many downstream proteolytic effects related to tissue remodeling, as described above, and *PLAU*-*PLAUR* have been linked to angiogenesis. In addition, *PLAUR* interacts with several membrane receptors, leading to the activation of multiple signaling cascades (phosphoinositide 3-kinase, ERK1/2, and mitogen-activated protein kinase), which leads to plasmin-independent effects that are important for normal airway cell functions, such as migration, proliferation, and adhesion.³²

The link between the PAS and structural changes in the airways *in vivo* has been clearly demonstrated in a recent study, showing that inhalation of *PLAU* in a murine model of asthma significantly attenuates subepithelial fibrosis and decreases AHR to methacholine.³³ We have also demonstrated that *PLAUR* is extensively expressed in both inflammatory and structural cells of the airways, including bronchial epithelial cells and ASM, and exists in multiple splice forms.³⁴ In addition to the genetic association between lung function decline in asthmatic subjects, we have also demonstrated that *PLAUR* is a susceptibility gene for the impairment of lung function in smokers.³⁵ As commented above, *ESR1* might influence plasmin generation, and a recent article has also implicated ADAM proteins in mediating *PLAUR* activation by using the ADAM inhibitors TNF- α protease inhibitor (TAPI)-0, and TAPI-1,³⁶ suggesting there might be functional relationship between the mechanisms underlying the associations with ADAM33, *ESR1*, and *PLAUR*.

It is important to note that several polymorphisms spanning *PLAU*³⁷ and *PAI1/SERPINE1*³⁸ have shown association with asthma susceptibility, and as for *PLAUR*, the asthma risk alleles within *SERPINE1* were associated with increased PAI-1 plasma levels.³⁹ These data suggest that genetic predisposition to the altered pathway activity might contribute to the development and progression of asthma; however, the contribution of *PLAU* and *SERPINE1* SNPs to airway remodeling and decline in lung function remains to be determined.

VEGF

VEGF is a critical angiogenic factor thought to lead to obstruction of the airways through alterations in inflammation, mucous metaplasia, smooth muscle hyperplasia, and parenchymal and vascular remodeling, as demonstrated in mice.⁴⁰ Importantly, increased levels of VEGF and its 2 receptors (VEGFR1/2) have been documented in asthmatic subjects in the airway mucosa.⁴¹ All 3 proteins were correlated with measures of remodeling, including vascularity.⁴¹ This and other studies led to the hypothesis that SNPs spanning *VEGF* might be important

susceptibility markers for airway remodeling in asthmatic subjects. *VEGF* SNP rs833058 (5' region) was associated with childhood asthma,²⁴ and rs4711750TT was associated with excess decline in FEV₁/FVC in asthmatic subjects followed for 4 years, indicating fixed airway obstruction.²⁴ This SNP is located in the 3' region of the *VEGF* gene, and the mechanism underlying this association remains to be resolved, although it might be hypothesized that this SNP influences expression levels of VEGF or is in LD with a causative SNP. Recently, it has been shown that IL-25 generated from T cells during airway inflammation can stimulate endothelial cells to induce VEGF and VEGF receptor expression through the phosphoinositide 3-kinase/Akt and ERK1/2-mitogen-activated protein kinase pathways, linking inflammation to airway remodeling in asthmatic subjects.⁴² It has been shown that VEGF can induce expression of ADAM33, leading to migration in cancer cell lines,⁴³ and a role for PLAUR in mediating VEGF-induced migration of vascular endothelial cells has been identified.⁴⁴

IMPLICATIONS FOR ASTHMA

Although the current evidence for a genetic contribution to airway remodeling is driven by candidate gene studies, there is compelling evidence that these genes (ie, *ADAM33*, *ESR1*, *PLAUR*, and *VEGF*) influence pathways and cell functions that are codependent and might be envisaged to culminate in airway remodeling, as suggested by excess decline in FEV₁ in carriers of "risk alleles." This interplay, particularly driven by proteolytic activity in the airways, might be a therapeutic opportunity if these complex codependencies can be further dissected at the DNA, RNA, protein, and cellular levels. Importantly, *ESR1*, *VEGF*, and *PLAUR* have been identified as having significantly altered mRNA expression in airway epithelial cells isolated from asthmatic subjects,¹² supporting the potential central role of the epithelium in remodeling processes.

LUNG FUNCTION GWA STUDIES

There have been several recent large-scale GWA studies to identify genetic determinants of lung function in the general population. A GWA study with 20,288 subjects of European descent (phase 1) and 32,184 or more additional subjects (phase 2) identified association with common variants in *TNSI*, *GSTCD*, and *HTR4* for FEV₁ and hedgehog interacting protein (*HHIP*), advanced glycosylation end-product (*AGER*), and *THSD4* for FEV₁/FVC ratio.⁴⁵ In parallel, a second study with 20,890 participants confirmed the associations within *HHIP*, *GSTCD*, *AGER*, and *HTR4* and suggested a role for additional loci: *GPR126*, *ADAM19*, *FAM13A*, *PTCHI*, and *PID1*.⁴⁶ These large-scale studies extended previous smaller studies that also identified *HHIP*.^{47,48} The basis of these associations remains to be resolved; for example, *HHIP* is a type I transmembrane glycoprotein with a potential role in lung morphogenesis, suggesting a mechanism involving lung development. The contribution of lung function genes to airway remodeling also remains to be resolved, although it has been demonstrated that *AGER*^{-/-} mice have age-related pulmonary fibrosis.⁴⁹ Initial data suggest common variants within these genes are not major determinants of asthma susceptibility.⁵⁰ However, associations with lung function within asthmatic populations have been reported for individual loci, such as *HHIP* and combinations of loci (*HHIP*, *PTCHI*, *FAM13A*, *PID1*, and

AGER).⁵¹ Thus these lung function loci might be at least in part involved in airway remodeling. Future studies should be directed to determine whether low lung function is the result of decreased lung function growth, progressive loss of lung function, or both.

CONCLUSIONS

Candidate gene studies have been useful in providing support for a genetic contribution to airway remodeling; however, there has been limited replication of these findings, except for *ADAM33*. The next few years will see the publication of GWA analyses in existing longitudinal asthma cohorts and, although insightful, will be limited to existing phenotypic measures. There is a compelling need for large-scale longitudinal studies examining multiple markers of airway remodeling in conjunction with a decline in FEV₁ to be used for GWA analyses. Furthermore, the association of SNPs with their expression (expression quantitative trait loci) in airway tissue might provide more insight into the functional role of these SNPs in patients with airway disease. Current therapeutic options for asthma do not adequately target airway-remodeling processes. As the genetic predisposition to airway remodeling is further defined, there is potential to identify new pathways and targets that might lead to the identification of susceptible subjects within the asthmatic population and new therapeutic targets to attenuate remodeling in these subjects.

REFERENCES

- Koppelman GH. Gene by environment interaction in asthma. *Curr Allergy Asthma Rep* 2006;6:103-11.
- Postma D, Kerkhof M, Boezen H, Koppelman G. Asthma and COPD: common genes, common environments? *Am J Respir Crit Care Med* 2011;183:1588-94.
- Holloway JW, Yang IA, Holgate ST. Genetics of allergic disease. *J Allergy Clin Immunol* 2010;125(suppl):S81-94.
- Tang MLK, Wilson JW, Stewart AG, Royce SG. Airway remodelling in asthma: current understanding and implications for future therapies. *Pharmacol Ther* 2006;112:474-88.
- Payne DNR, Rogers AV, Adelroth E, Bandi V, Guntupalli KK, Bush A, et al. Early thickening of the reticular basement membrane in children with difficult asthma. *Am J Respir Crit Care Med* 2003;167:78-82.
- 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.
- Holgate ST. Epithelium dysfunction in asthma. *J Allergy Clin Immunol* 2007;120:1233-46.
- Shinagawa K, Kojima M. Mouse model of airway remodeling: strain differences. *Am J Respir Crit Care Med* 2003;168:959-67.
- Antunes MA, Abreu SC, Damasceno-Rodrigues NR, Parra ER, Capelozzi VL, Pinart M, et al. Different strains of mice present distinct lung tissue mechanics and extracellular matrix composition in a model of chronic allergic asthma. *Respir Physiol Neurobiol* 2009;165:202-7.
- Hubert HB, Fabsitz RR, Feinleib M, Gwinn C. Genetic and environmental influences on pulmonary function in adult twins. *Am Rev Respir Dis* 1982;125:409-15.
- Hopp RJ, Bewtra AK, Watt GD, Nair NM, Townley RG. Genetic analysis of allergic disease in twins. *J Allergy Clin Immunol* 1984;73:265-70.
- Kicic A, Hallstrand TS, Sutanto EN, Stevens PT, Kobor MS, Taplin C, et al. Decreased fibronectin production significantly contributes to dysregulated repair of asthmatic epithelium. *Am J Respir Crit Care Med* 2010;181:889-98.
- Wenzel SE, Balzar S, Ampleford E, Hawkins GA, Busse WW, Calhoun WJ, et al. IL4R alpha mutations are associated with asthma exacerbations and mast cell/IgE expression. *Am J Respir Crit Care Med* 2007;175:570-6.
- Chetta A, Foresi A, Del Donno M, Bertorelli G, Pesci A, Olivieri D. Airways remodeling is a distinctive feature of asthma and is related to severity of disease. *Chest* 1997;111:852-7.
- Boulet L, Belanger M, Carrier G. Airway responsiveness and bronchial-wall thickness in asthma with or without fixed airflow obstruction. *Am J Respir Crit Care Med* 1995;152:865-71.
- Tsurikisawa N, Oshikata C, Tsuburai T, Saito H, Sekiya K, Tanimoto H, et al. Bronchial hyperresponsiveness to histamine correlates with airway remodelling in adults with asthma. *Respir Med* 2010;104:1271-7.

17. Lange P, Parner J, Vestbo J, Schnohr P, Jensen G. A 15-year follow-up study of ventilatory function in adults with asthma. *N Engl J Med* 1998;339:1194-200.
18. James AL, Palmer LJ, Kicic E, Maxwell PS, Lagan SE, Ryan GF, et al. Decline in lung function in the Busselton Health Study: the effects of asthma and cigarette smoking. *Am J Respir Crit Care Med* 2005;171:109-14.
19. Dijkstra A, Vonk JM, Jongepier H, Koppelman GH, Schouten JP, ten Hacken NHT, et al. Lung function decline in asthma: association with inhaled corticosteroids, smoking and sex. *Thorax* 2006;61:105-10.
20. Vonk JM, Jongepier H, Panhuysen CIM, Schouten JP, Bleecker ER, Postma DS. Risk factors associated with the presence of irreversible airflow limitation and reduced transfer coefficient in patients with asthma after 26 years of follow up. *Thorax* 2003;58:322-7.
21. Jongepier H, Boezen HM, Dijkstra A, Howard TD, Vonk JM, Koppelman GH, et al. Polymorphisms of the ADAM33 gene are associated with accelerated lung function decline in asthma. *Clin Exp Allergy* 2004;34:757-60.
22. Dijkstra A, Howard TD, Vonk JM, Ampleford EJ, Lange LA, Bleecker ER, et al. Estrogen receptor 1 polymorphisms are associated with airway hyperresponsiveness and lung function decline, particularly in female subjects with asthma. *J Allergy Clin Immunol* 2006;117:604-11.
23. Barton SJ, Koppelman GH, Vonk JM, Browning CA, Nolte IM, Stewart CE, et al. PLAUR polymorphisms are associated with asthma, PLAUR levels, and lung function decline. *J Allergy Clin Immunol* 2009;123:1391-400, e17.
24. Sharma S, Murphy AJ, Soto-Quiros ME, Avila L, Klanderman BJ, Sylvia JS, et al. Association of VEGF polymorphisms with childhood asthma, lung function and airway responsiveness. *Eur Respir J* 2009;33:1287-94.
25. van Diemen CC, Postma DS, Vonk JM, Bruinenberg M, Schouten JP, Boezen HM. A disintegrin and metalloprotease 33 polymorphisms and lung function decline in the general population. *Am J Respir Crit Care Med* 2005;172:329-33.
26. Vonk JM, Postma DS, Maarsingh H, Bruinenberg M, Koppelman GH, Meurs H. Arginase 1 and arginase 2 variations associate with asthma, asthma severity and beta2 agonist and steroid response. *Pharmacogenet Genomics* 2010;20:179-86.
27. Van Eerdewegh P, Little RD, Dupuis J, Del Mastro RG, Falls K, Simon J, et al. Association of the ADAM33 gene with asthma and bronchial hyperresponsiveness. *Nature* 2002;418:426-30.
28. Holgate ST, Davies DE, Powell RM, Holloway JW. ADAM33: a newly identified protease involved in airway remodelling. *Pulm Pharmacol Ther* 2006;19:3-11.
29. Puxeddu I, Pang YY, Harvey A, Haitchi HM, Nicholas B, Yoshisue H, et al. The soluble form of a disintegrin and metalloprotease 33 promotes angiogenesis: implications for airway remodeling in asthma. *J Allergy Clin Immunol* 2008;121:1400-6, e1-4.
30. Rossouw J, Bray P, Liu J, Kooperberg C, Hsia J, Lewis C, et al. Estrogen receptor polymorphisms and the vascular effects of hormone therapy. *Arterioscler Thromb Vasc Biol* 2011;31:464-9.
31. Flores-Delgado G, Bringas P, Buckley S, Anderson KD, Warburton D. Nongenomic estrogen action in human lung myofibroblasts. *Biochem Biophys Res Commun* 2001;283:661-7.
32. Blasi F, Carmeliet P. uPAR: a versatile signalling orchestrator. *Nat Rev Mol Cell Biol* 2002;3:932-43.
33. Kuramoto E, Nishiuma T, Kobayashi K, Yamamoto M, Kono Y, Funada Y, et al. Inhalation of urokinase-type plasminogen activator reduces airway remodeling in a murine asthma model. *Am J Physiol Lung Cell Mol Physiol* 2009;296:L337-46.
34. Stewart CE, Sayers I. Characterisation of urokinase plasminogen activator receptor variants in human airway and peripheral cells. *BMC Mol Biol* 2009;10:75.
35. Stewart CE, Hall IP, Parker SG, Moffat MF, Wardlaw AJ, Connolly MJ, et al. PLAUR polymorphisms and lung function in UK smokers. *BMC Med Genet* 2009;10:112.
36. Bakken AM, Protack CD, Roztocil E, Nicholl SM, Davies MG. Cell migration in response to the amino-terminal fragment of urokinase requires epidermal growth factor receptor activation through an ADAM-mediated mechanism. *J Vasc Surg* 2009;49:1296-303.
37. Begin P, Tremblay K, Daley D, Lemire M, Claveau S, Salesse C, et al. Association of urokinase-type plasminogen activator with asthma and atopy. *Am J Respir Crit Care Med* 2007;175:1109-16.
38. Cho SH, Hall IP, Wheatley A, Dewar J, Abraha D, Del Mundo J, et al. Possible role of the 4G/5G polymorphism of the plasminogen activator inhibitor 1 gene in the development of asthma. *J Allergy Clin Immunol* 2001;108:212-4.
39. Pampuch A, Kowal K, Bodzenta-Lukaszyk A, Di Castelnuovo A, Chyczewski L, Donati MB, et al. The -675 4G/5G plasminogen activator inhibitor-1 promoter polymorphism in house dust mite-sensitive allergic asthma patients. *Allergy* 2006;61:234-8.
40. Lee CG, Link H, Baluk P, Homer RJ, Chapoval S, Bhandari V, et al. Vascular endothelial growth factor (VEGF) induces remodeling and enhances TH2-mediated sensitization and inflammation in the lung. *Nat Med* 2004;10:1095-103.
41. Hoshino M, Nakamura Y, Hamid QA. Gene expression of vascular endothelial growth factor and its receptors and angiogenesis in bronchial asthma. *J Allergy Clin Immunol* 2001;107:1034-8.
42. Corrigan C, Wang W, Meng Q, Fang C, Wu H, Reay V, et al. T-helper cell type 2 (Th2) memory T cell-potentiating cytokine IL-25 has the potential to promote angiogenesis in asthma. *Proc Natl Acad Sci U S A* 2011;25:1579-84.
43. Kim K-E, Song H, Hahm C, Yoon SY, Park S, Lee H-r, et al. Expression of ADAM33 is a novel regulatory mechanism in IL-18-secreted process in gastric cancer. *J Immunol* 2009;182:3548-55.
44. Prager GW, Breuss JM, Steurer S, Olcaydu D, Mihaly J, Brunner PM, et al. Vascular endothelial growth factor receptor-2-induced initial endothelial cell migration depends on the presence of the urokinase receptor. *Circ Res* 2004;94:1562-70.
45. Repapi E, Sayers I, Wain LV, Burton PR, Johnson T, Obeidat M, et al. Genome-wide association study identifies five loci associated with lung function. *Nat Genet* 2010;42:36-44.
46. Hancock DB, Eijgelsheim M, Wilk JB, Gharib SA, Loehr LR, Marcianti KD, et al. Meta-analyses of genome-wide association studies identify multiple loci associated with pulmonary function. *Nat Genet* 2010;42:45-52.
47. Wilk JB, Walter RE, Laramie JM, Gottlieb DJ, O'Connor GT. Framingham Heart Study genome-wide association: results for pulmonary function measures. *BMC Med Genet* 2007;8(suppl 1):S8.
48. Wilk JB, Chen T-H, Gottlieb DJ, Walter RE, Nagle MW, Brandler BJ, et al. A genome-wide association study of pulmonary function measures in the Framingham Heart Study. *PLoS Genet* 2009;5:e1000429.
49. Englert JM, Hanford LE, Kaminski N, Tobolewski JM, Tan RJ, Fattman CL, et al. A role for the receptor for advanced glycation end products in idiopathic pulmonary fibrosis. *Am J Pathol* 2008;172:583-91.
50. Moffatt MF, Gut IG, Demenais F, Strachan DP, Bouzigon E, Heath S, et al. A large-scale, consortium-based genomewide association study of asthma. *N Engl J Med* 2010;363:1211-21.
51. Li X, Howard T, Moore W, Ampleford E, Li H, Busse W, et al. Importance of hedgehog interacting protein and other lung function genes in asthma. *J Allergy Clin Immunol* 2011;127:1457-65.