

Integrin $\beta 3$ genotype influences asthma and allergy phenotypes in the first 6 years of life

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Background: The integrin $\beta 3$ gene (*ITGB3*) encodes a subunit of the platelet and monocyte-specific fibrinogen receptor and the widely expressed vitronectin receptor, which have diverse roles in cell migration, adhesion, and signaling. Previous work from our laboratory reported associations between single nucleotide polymorphisms (SNPs) in *ITGB3* and asthma and allergic sensitization in 4 populations.

Objective: To examine whether SNPs in *ITGB3* are associated with the development of asthma and allergic phenotypes in early life.

Methods: We typed 13 SNPs in 206 children participating in a birth cohort study and tested for associations with asthma and allergy phenotypes in the first 6 years of life.

Results: Our study revealed significant associations between SNPs in *ITGB3* and asthma, wheezing, and IgE levels, suggesting an early role for this gene in the development of asthma and allergy. In particular, SNPs at the 3' end of the gene were significantly associated with IgE levels beginning at 1 year of age, whereas a SNP in intron 1 showed significant interaction effects with viral respiratory illness in infancy on asthma susceptibility.

Conclusion: Our results suggest that genetic variation in *ITGB3* contributes to asthma susceptibility and allergic sensitization, and that the effects of this gene begin early in life. Similar to our earlier study, different SNPs in the gene are associated with asthma and IgE.

Clinical implications: *ITGB3* may play an important role in the development of asthma and allergy and may represent a potential therapeutic target for the treatment of these disorders. (*J Allergy Clin Immunol* 2007;119:1423-9.)

Key words: Asthma, allergy, noncoding variation, integrin, viral respiratory illness

Abbreviations used

ADAM33: A disintegrin and matrix metalloprotease
BHR: Bronchial hyperresponsiveness
COAST: Childhood Onset of ASThma
GEI: Gene-environment interaction
ITGB3: Integrin $\beta 3$
LD: Linkage disequilibrium
SNP: Single nucleotide polymorphism
UTR: Untranslated region
VRI: Viral respiratory illness

Asthma is a heterogeneous respiratory disease characterized by reversible airway obstruction and bronchial hyperresponsiveness (BHR). Susceptibility to asthma likely involves interactions between genotypes at many loci and environmental exposures, particularly those that occur early in life (see review¹). Of the > 100 genes that have been implicated in asthma or allergy susceptibility (see reviews^{2,3}), relatively few have been consistently replicated in multiple populations. Even among the few consistently replicated genes, associations are infrequently observed with the same phenotype or with the same polymorphism, indicating that genetic susceptibility is complex and likely to be context-dependent.⁴⁻⁶ Because of the prominent roles inflammation and immune dysregulation play in asthma pathogenesis, the overwhelming majority of asthma or atopy genes investigated to date are involved in inflammation and immunoregulation pathways. However, recent studies have reported associations with genes encoding integrins or related genes, such as a disintegrin and matrix metalloprotease (*ADAM33*),⁷ P-selectin (*SELP*),^{8,9} and integrin $\beta 3$ (*ITGB3*),¹⁰ implicating these pathways in the pathogenesis of asthma and allergic disease. Moreover, ADAM family members, including *ADAM33*, selectively regulate integrin-mediated cell migration,¹¹ suggesting that genetic interactions between genes in these pathways may also be important.

ITGB3 encodes a subunit of the platelet and monocyte-specific fibrinogen receptor and the widely expressed vitronectin receptor. These receptors have diverse roles, including cell migration,¹² cell adhesion,¹³ and signaling.¹⁴ In our earlier study, we tested 19 single nucleotide polymorphisms (SNPs) in this gene for association with BHR and atopy in the Hutterites, a religious isolate of European descent that has been the focus of asthma genetic studies in our laboratory.^{10,15-19} Five SNPs were associated with BHR and 7 with allergic sensitization in

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Supported by P01 HL70831; E.E.T. is supported by T32 HL007605.

Disclosure of potential conflict of interest: R. F. Lemanske has consulting arrangements with Merck, GlaxoSmithKline, Aventis, AstraZeneca, and Novartis; has received grant support from the National Institutes of Allergy and Infectious Diseases and the National Heart, Lung, and Blood Institute; and is on the speakers' bureau for Merck, GlaxoSmithKline, and AstraZeneca. C. Ober has received grant support from the National Institutes of Health. The rest of the authors have declared that they have no conflict of interest.

Received for publication January 2, 2007; revised March 20, 2007; accepted for publication March 26, 2007.

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0091-6749/\$32.00

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doi:10.1016/j.jaci.2007.03.029

the Hutterites at the $P < .001$ significance level. We then genotyped a subset of these SNPs in African American and white asthma cases and controls from Chicago, and white asthma cases and controls from Madison, Wis, and identified associations ($P < .05$) with multiple SNPs for both phenotypes in these samples, although the associated SNPs and alleles differed between them. Only 1 coding variant (Leu33Pro) is present in this gene, but this was never the most significantly associated SNP. Because there is little haplotype structure in this gene (ie, few SNPs were in linkage disequilibrium [LD]; see Fig 2 in Weiss et al²⁰), it was unlikely that the associated SNPs were in LD with a shared susceptibility variant. The association with noncoding variation further suggested that variation in gene expression levels may influence disease susceptibility, as discussed.^{20,21} Thus, although we concluded that variation in *ITGB3* influences risk for asthma and atopy, the link between genotype and phenotype was not at all clear. Moreover, this pattern of replication in which different SNPs or different alleles are associated in different populations suggested more complex models of susceptibility, such as gene-environment interactions (GEIs).⁴⁻⁶

The sample from Madison, Wis, consisted of 412 unrelated white adults who were the parents of children participants in a birth cohort study, called the Childhood Onset of ASThma (COAST) Study.²² For this study, parents were recruited in their 9th month of pregnancy and were eligible for participation if at least 1 parent had atopy (assessed by skin prick test) and/or asthma (assessed by current or past medication use or doctor's diagnosis), as previously described.²² Twelve SNPs across the gene were associated with asthma or allergic sensitization in the COAST parents ($P < .05$).¹⁰ In the current study, we genotyped SNPs in the COAST children and examined the effects of this gene on the natural history of asthma and allergy and GEIs in the first 6 years of life.

METHODS

Population

The COAST study has been described in detail elsewhere.²²⁻²⁶ Our genetic studies focus on 206 unrelated white children whose parents gave consent for genetic studies, and include 1 each of 9 twin pairs, as previously described.^{23,27} Because our previous study identified associations with asthma or BHR and allergic sensitization, we focus here on related phenotypes measured in the first 6 years: total IgE at ages 1, 2, 3, 5, and 6 years (age 4 years measurements are not available), RAST and skin prick test to aeroallergens at age 5 or 6 years, wheezing at age 5 years, and asthma at age 6 years. At age 6 years, asthma was diagnosed in the COAST children who met at least 1 of the following criteria: (1) doctor-diagnosed asthma; (2) doctor-prescribed albuterol use for coughing or wheezing episodes more than once between 60 and 72 months of age; (3) daily use of controller medication; (4) implemented a step-up plan as prescribed by an MD including the use of albuterol or short-term use of ICS during illness; and/or (5) use of prescribed prednisone for asthma exacerbation.

Selection of SNPs and genotyping

Ten SNPs that were genotyped in the COAST parents were selected for genotype studies in their children. The initial SNP set was

spaced ~5 kb across the gene, spanning from 5 kb upstream to 5 kb downstream of the *ITGB3* exons. After analysis with LdSelect²⁸ to determine maximal SNP coverage (data not shown), 3 additional SNPs (rs2056131, rs2015049, and rs3851806) were selected for genotyping. Last, we selected for genotyping an intron 1 SNP (rs2015049), which is in perfect LD with the 5' SNP, rs2317385, genotyped in our earlier study (http://www.hapmap.org/cgi-perl/gbrowse/hapmap_B35/). The resulting 13 SNPs serve as a set of markers that could be used for genotyping *ITGB3* in other white populations, minimizing redundancy and capturing genetic variation occurring at 10% or greater. The LD structure of *ITGB3* in 3 populations was shown in a previous publication by our group.²¹

Nine of the original 10 SNPs were genotyped by DNAPrint, Inc (Sarasota, Fla; <http://www.dnaprint.com/>); the remaining SNP (rs15908) and the 3 new SNPs were genotyped by TaqMan SNP Genotyping Assays (Applied Biosystems, Foster City, Calif). As in our earlier study,¹⁰ a chimpanzee sequence (NW_118536.1) was used to define the ancestral state of each allele.

Statistical analyses

Hardy-Weinberg equilibrium was evaluated by the χ^2 test. Haplotypes were inferred by using PHASE2.^{29,30} Association tests were performed by using χ^2 tests for binary traits and the nonparametric Kruskal-Wallis ANOVA test for quantitative traits (IgE). Because our previous hypothesis was that SNPs in *ITGB3* are associated with asthma and atopic phenotypes, we did not correct for multiple testing.

We also examined GEIs with "number of moderate to severe respiratory viral illnesses" in infancy by including this variable as a covariate in our analyses, as in our previous studies.^{23,24} We selected this exposure because viral respiratory illness (VRI) in infancy was the most significant predictor of wheezing at age 3 years³¹ and remained a significant predictor of asthma at age 6 years (R.F. Lemanske Jr, unpublished data, 2007) in COAST children.

All P values were determined by using a permutation-based method that takes into account small sizes, as described previously.²³

RESULTS

A schematic illustration of the *ITGB3* gene and the SNPs typed in our study are shown in Table I. All SNPs were in Hardy-Weinberg equilibrium ($P > .10$). The 13 SNPs include 3 exonic (2 synonymous and 1 nonsynonymous), 8 intronic, 1 SNP in the 3' untranslated region (UTR), and 1 downstream of *ITGB3*.

We first examined main effects of these SNPs (Table I). Two SNPs were associated with IgE levels (rs2317676, rs2317677), 3 were associated with wheezing at age 5 years (rs5918, rs3760372, rs2317677), and 2 were associated with asthma at age 6 years (rs5918, rs4634). Four of the 5 SNPs associated in the COAST children were also associated with asthma and/or atopy in the parents, and in each case the same allele was associated in both parents and children. Two of those SNPs (rs5918, rs2317677) were each associated with 2 phenotypes (rs5918 with wheezing at age 5 years and asthma at age 6 years; rs2317677 with IgE at ages 2, 3, and 5 years and wheezing at age 5 years). SNP rs5918 is the only nonsynonymous SNP in the gene (Leu33Pro). The Pro allele was modestly associated with wheezing at age 5 years and asthma at age 6 years in the children, and with allergic sensitization in their parents.¹⁰ SNP rs2317677 is located 3' of the gene,

TABLE I. Schematic illustration of the *ITGB3* gene and 13 SNPs genotyped in the COAST children with *P* values < .10 for SNPs with main effects or < .01 for interaction effects

	rs 2015049	rs 2056131	rs 3851806	rs 2015729	rs 1000232	rs 5918 (L33P)	rs 2292864	rs 15908 (V381V)	rs4634 (R515R)	rs 999323	rs 3760372	rs 2317676	rs 2317677
Genotyped (N)	194	191	194	197	199	202	200	180	187	194	199	203	197
Parents*	NT	NT	NT	AS	AL	AL	AS	AS	AL	AL	AL	AS	AL
SNP†	A/G	C/T	C/G	A/G	C/T	A/G	A/G	A/C	A/G	A/G	A/G	G/A	A/G
Derived allele frequency	0.78	0.67	0.82	0.61	0.39	0.14	0.12	0.64	0.3	0.32	0.31	0.06	0.61
Main effects													
IgE at 1 y (N = 202)												.004	.081
IgE at 2 y (N = 190)												.010	.019
IgE at 3 y (N = 178)												.036	.006
IgE at 5 y (N = 155)												.038	.020
IgE at 6 y (N = 139)												.023	
Wheezing at age 5 y (N = 189)						.049					.001		.027
Asthma at age 6 y (N = 185)						.034			.044				
Interaction effects with respiratory viral infection in infancy (interaction <i>P</i> value is shown)‡													
Wheezing at age 5 y			.009										
Asthma at age 6 y			.008										

*SNPs that were associated with asthma (AS) or allergic sensitization (AL) in the parents are shown on a shaded background (NT, not typed).

†The associated alleles in the main effects analysis in the children are *underlined*.

‡Only phenotypes with significant interactions are shown.

and the minor (ancestral) allele at this SNP was associated with IgE at ages 2 to 5 years and wheezing at age 5 years in the children, and with allergic sensitization in the parents.¹⁰

Two SNPs (rs2317677 and rs2317676) were associated with IgE levels at 1 to 6 years of age (Fig 1). Genotype-specific differences in IgE levels are apparent by 1 year of age and remain different over the period of the next 5 years, even though mean IgE levels increased from age 1 to age 6 years in all genotype groups. At both SNPs, the ancestral alleles are associated with increased IgE levels, which is the common allele at rs2317677 (frequency 0.94) and the minor allele at rs2317677 (frequency 0.39). Haplotypes carrying the ancestral alleles at both rs2317676 and rs2317677 account for 32% of the total chromosomes in our sample. However, these alleles tag a heterogeneous group of haplotypes on which these 2 SNPs are the only shared variants (Fig 2). The 3 haplotype groups tagged by these 2 SNPs (Fig 2) showed the same pattern of association with IgE levels as the individual SNPs, but with overall larger *P* values than those observed with rs2317677. The latter suggests that these SNPs do not tag a haplotype with additional unknown/untyped variation that influences IgE levels and that the 2 SNPs independently influence IgE levels.

We next examined GEIs between SNPs in *ITGB3* and moderate to severe respiratory illness in infancy, the

most significant exposure in infancy for subsequent development of wheezing and asthma. The mean number of viral infections in infancy in the COAST children was 1.442 (SD, 1.416). In this analysis, we considered interaction effects between the exposure and each of 12 SNPs on the 7 phenotypes shown in Table I. We did not include rs2317676 in this analysis because there were too few counts as a result of the low minor allele frequency (0.06) for meaningful analyses. Among the remaining 84 GEIs examined, 2 had *P* values < .01. Surprisingly, however, these were both with a SNP (rs3851806) in intron 1 that did not show main effect associations with any of the phenotypes (Table I). SNP rs3851806 showed significant interaction effects with both wheezing at age 5 years and asthma at age 6 years (Fig 3). The relationship between number of viral respiratory infections in infancy and wheezing at age 5 years (Fig 3, A) and asthma at age 6 years (Fig 3, B) was present only among children with the GG genotype. Among GG children who had 3 or more viral infections, > 40% (11/26) wheezed at age 5 years and > 50% (13/25) had asthma at age 6 years compared with < 10% (1/11) and < 30% (3/11), respectively, of children with the GC or CC genotype. In fact, among the latter group of children, the risk for wheeze and asthma did not change with increasing number of viral infections (Fig 3). Although the number of GC or CC children with 3

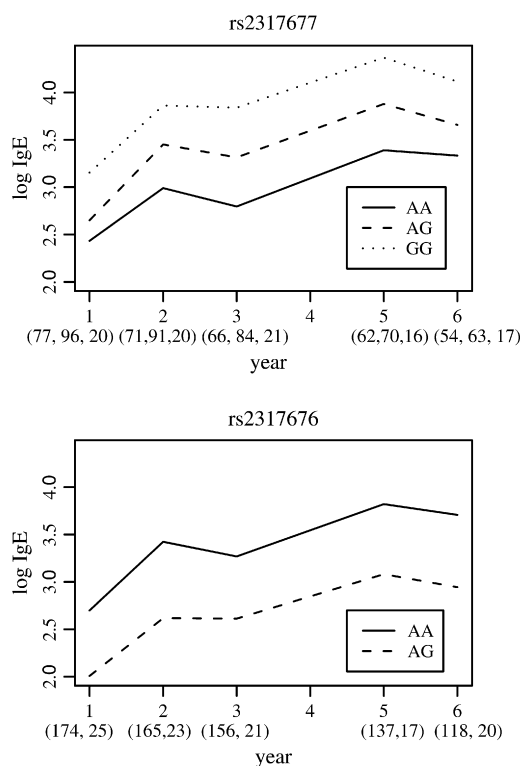


FIG 1. Increase in IgE level through year 6 by *ITGB3* genotype ($.007 < P < .05$). IgE levels were not available for year 4. Sample sizes for each genotype are shown along the x-axis in the order shown in the legend.

or more viral infections in infancy is small ($N = 11$), the trend is striking given that only 1 of these children wheezed at age 5 years and only 3 developed asthma by age 6 years, raising the possibility that viral respiratory infections in infancy may protect against subsequent development of asthma in children with a C allele at this SNP.

DISCUSSION

On the basis of our previous report of associations of *ITGB3* SNPs with asthma-related and atopy-related phenotypes in multiple populations,¹⁰ we expanded our investigation to include the children of 1 of these populations and to explore genotype-specific effects on the natural history of asthma and allergy. Our survey of 13 SNPs in *ITGB3* in 206 children revealed associations with IgE levels that were evident at 1 year of age and with wheezing at age 5 years and asthma at age 6 years. Thus, this study not only further suggests a role for this gene in asthma pathogenesis but also indicates that the effects of this gene begin in the first year of life. Moreover, the patterns of association revealed by this and our previous study¹⁰ suggest that multiple SNPs contribute to susceptibility, perhaps at different stages of development, and that genotype-specific risks may be context-dependent, interacting with exposures in early life. In the COAST children,

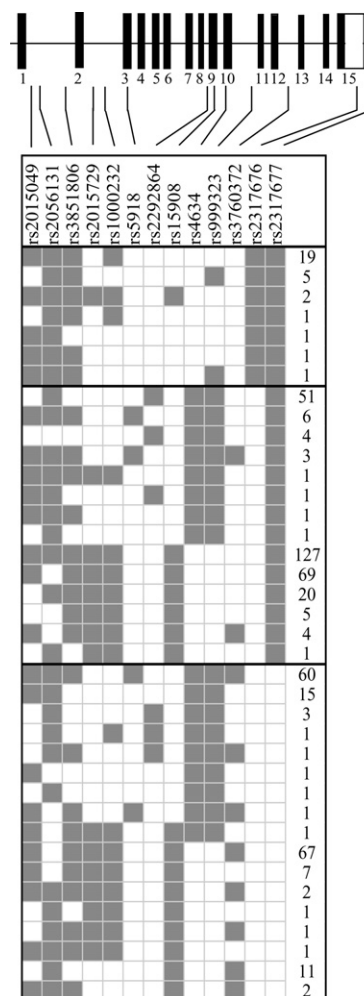


FIG 2. Visual representation of haplotypes at *ITGB3*. A schematic illustration of the *ITGB3* gene is shown at the top of the figure. White boxes indicate ancestral alleles and gray derived alleles. The number of each chromosome/haplotype observed is shown in the far right column. Haplotypes are sorted by the IgE-associated SNPs (rs2317676 and rs2317677).

SNPs in the 3' region of the gene had main effects on IgE levels, whereas a SNP in intron 1 interacted with VRIs in infancy to influence risk for the subsequent development wheezing illnesses and asthma.

The 2 SNPs at the 3' end of the gene that are associated with IgE level in COAST children (Fig 1) were also associated with asthma or allergic sensitization in their parents and in an unrelated case-control sample of Chicago white subjects,¹⁰ suggesting that either the SNPs themselves or other untyped SNPs in LD with them influence susceptibility. Their location in the 3' region (1 in the 3' UTR), and the absence of any coding SNPs in LD with these SNPs,²⁰ suggest that the associated SNPs influence expression level or mRNA stability. In fact, SNPs in the 3' UTR can modulate mRNA or protein expression levels through a variety of mechanisms.³²⁻³⁶ Moreover, integrins have been implicated in cellular interactions necessary for IgE synthesis³⁷ and in asthma pathogenesis.³⁸

In particular, $\beta 3$ -integrin *ITGB3*-deficient mice show elevated *TGF- $\beta 1$* signaling and enhanced/accelerated wound healing.³⁸ *TGF- $\beta 1$* is highly expressed in the respiratory tract and is a negative regulator of airway inflammation and hyperresponsiveness.^{39,40} *TGF- $\beta 1$* is generally found to be more highly expressed in the airways of patients with asthma compared with controls without asthma,⁴¹ perhaps reflecting remodeling processes of asthmatic airways.⁴² Therefore, SNPs that reduce expression of *ITGB3* could result in elevated *TGF- $\beta 1$* levels in the airway and contribute toward asthma pathogenesis, as our data suggest.

The single coding SNP in this gene, Leu33Pro, defines the platelet antigen P1^{A1/A2} that influences receptor signaling in human platelets, with the Pro allele conferring enhanced signaling.¹⁴ This SNP was modestly associated with asthma in the COAST children and showed a similarly modest association with allergic sensitization in COAST parents and in Hutterites.¹⁰ It is possible, therefore, that the signaling properties mediated by this SNP could affect downstream targets of *ITGB3* that influence asthma susceptibility, or could influence susceptibility directly through some other mechanism. In either case, this SNP was consistently among the less associated SNPs in both studies and does not account for a significant proportion of the genetic risk associated with this gene.

According to HapMap data, 2 of the 3 most significant SNPs in *ITGB3* (rs3851806 and rs2317676; rs2317677 has not been typed) are each in LD with surrounding SNPs; it is possible, therefore, that the results we observe are merely a result of their association with the real risk variants. The patterns of LD, however, would suggest that the true risk variants, if not those reported here, are nonetheless within *ITGB3*, because the intron 1 SNP (rs3851806) is primarily in strong LD with other intron 1 variants, and the 3' UTR SNP (rs2317676) lies in a block of LD with other markers at the 3' end of the gene (data not shown). In addition, the 2 highly significant 3' SNPs reported in this study are not in LD with one another, suggesting a role for 3' variation in *ITGB3* regulation regardless.

Examination of an important environmental risk factor in infancy in the COAST cohort revealed additional complexities of association. SNPs in intron 1 showed significant GEIs with the number of VRIs in the first year (Table I and Fig 3). These SNPs were not included in our earlier study, so we do not know if they are associated with asthma or allergy in the 4 samples included in that study. COAST children with 1 or 2 copies of the C allele at SNP rs3851806 were protected against the effects of viral illnesses on asthma and wheezing risks, whereas 3 or more VRIs was a significant risk factor for wheezing and asthma among children with the GG genotype. This GEI is similar to those reported for other exposures, where genotype-specific risks for atopic phenotypes differ by environmental exposures.⁴³⁻⁴⁵ We previously reported GEIs between daycare exposure in the first 6 months of life and first year immune phenotypes in COAST children.²³ Significant GEIs with SNPs in the IL-4 receptor α (*IL4RA*) gene and IFN- γ responses and the *FCERB1* gene and IL-5 responses

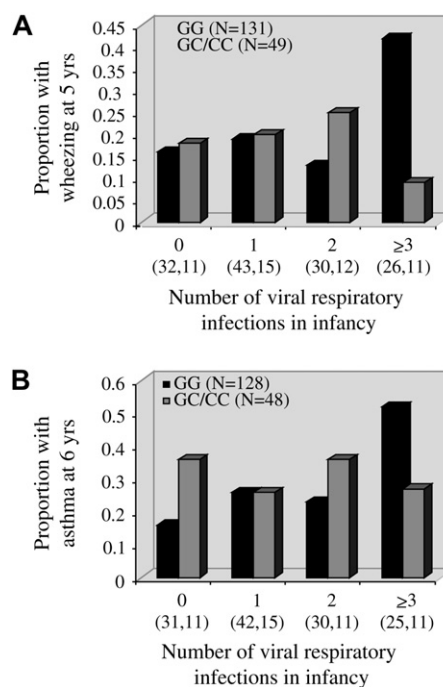


FIG 3. Interaction plots for rs3851806 with number of moderate to severe VRIs in infancy. **A**, Wheezing at age 5 years. **B**, Asthma at age 6 years. Interaction *P* values = .009 and .008, respectively. Sample sizes for each genotype group appear in the legend; sample sizes for each year on the x-axis (GG, GC/CC). Sample sizes are reported in the text.

were nearly completely accounted for by the increased number of VRIs among children attending daycare. Therefore, genotype-specific risks at loci that influence T_H1 (IFN- γ) and T_H2 (IL-5) responsiveness in mononuclear cells at 1 year of age differ by exposure to moderate to severe VRIs in infancy, and may further interact with *ITGB3* genotype to determine subsequent risk for asthma. Although the size of the COAST cohort is too small to examine these higher levels of interaction, it would not be surprising if such interactions exist and contribute to asthma susceptibility. Last, interactions with VRIs in infancy, or with other relevant environmental exposures, could account for associations with different *ITGB3* alleles in different populations, as we previously reported.¹⁰

In summary, our results further support a role for *ITGB3* in asthma susceptibility, and extend our earlier findings to indicate that the effects of this gene on the development of atopic phenotypes begin in the first year of life. Furthermore, SNPs in this gene show interaction effects with VRIs in infancy on asthma risk. The fact that different SNPs throughout the gene are associated with IgE levels, asthma, and with viral interactions suggest that the effects of this gene act through multiple mechanisms, which likely influence gene regulation and/or expression. Regardless of the mechanism, however, these studies implicate integrins in asthma pathogenesis and response to viral respiratory infections in infancy, as previously suggested.^{10,46,47}

We thank the COAST families and staff in Madison, Wis. The COAST project would not be possible without the participation and cooperation of the following hospitals and organizations: Associated Physicians LLP, Dean Medical Center, Group Health Cooperative, Physicians Plus Group Practices, University of Wisconsin Health, Prairie Clinic Sauk City, Reedsburg Physician Group, St Mary's Hospital Medical Center, Meriter Hospital, St Clare Hospital and Health Care Services, Sauk Prairie Memorial Hospital, Reedsburg Area Medical Center, and Fort Atkinson Memorial Health Care Services.

REFERENCES

- Ober C. Perspectives on the past decade of asthma genetics. *J Allergy Clin Immunol* 2005;116:274-8.
- Bosse Y, Hudson TJ. Toward a comprehensive set of asthma susceptibility genes. *Annu Rev Med* 2007;58:171-84.
- Ober C, Hoffjan S. Asthma genetics 2006: the long and winding road to gene discovery. *Genes Immun* 2006;7:95-100.
- Ober C, Thompson EE. Rethinking genetic models of asthma: the role of environmental modifiers. *Curr Opin Immunol* 2005;17:670-8.
- Martinez FD. Gene-environment interactions in asthma and allergies: a new paradigm to understand disease causation. *Immunol Allergy Clin North Am* 2005;25:709-21.
- Vercelli D, Martinez FD. The Faustian bargain of genetic association studies: bigger might not be better, or at least it might not be good enough. *J Allergy Clin Immunol* 2006;117:1303-5.
- 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.
- Bourgain C, Hoffjan S, Nicolae R, Newman D, Steiner L, Walker K, et al. Novel case-control test in a founder population identifies P-selectin as an atopy-susceptibility locus. *Am J Hum Genet* 2003;73:612-26.
- Woltmann G, McNulty CA, Dewson G, Symon FA, Wardlaw AJ. Interleukin-13 induces PSGL-1/P-selectin-dependent adhesion of eosinophils, but not neutrophils, to human umbilical vein endothelial cells under flow. *Blood* 2000;95:3146-52.
- Weiss LA, Lester LA, Gern JE, Wolf RL, Parry R, Lemanske RF, et al. Variation in ITGB3 is associated with asthma and sensitization to mold allergen in four populations. *Am J Respir Crit Care Med* 2005;172:67-73.
- Huang J, Bridges LC, White JM. Selective modulation of integrin-mediated cell migration by distinct ADAM family members. *Mol Biol Cell* 2005;16:4982-91.
- Ridley AJ, Schwartz MA, Burridge K, Firtel RA, Ginsberg MH, Borisy G, et al. Cell migration: integrating signals from front to back. *Science* 2003;302:1704-9.
- Thompson RD, Wakelin MW, Larbi KY, Dewar A, Asimakopoulos G, Horton MA, et al. Divergent effects of platelet-endothelial cell adhesion molecule-1 and beta 3 integrin blockade on leukocyte transmigration in vivo. *J Immunol* 2000;165:426-34.
- Vijayan KV, Liu Y, Sun W, Ito M, Bray PF. The Pro33 isoform of integrin beta3 enhances outside-in signaling in human platelets by regulating the activation of serine/threonine phosphatases. *J Biol Chem* 2005;280:21756-62.
- Chan A, Newman DL, Shon AM, Schneider DH, Kuldaneck S, Ober C. Variation in the type I interferon gene cluster on 9p21 influences susceptibility to asthma and atopy. *Genes Immun* 2006;7:169-78.
- Ober C, Leavitt SA, Salenko A, Howard TD, Hoki DM, Daniel R, et al. Variation in the interleukin 4-receptor alpha gene confers susceptibility to asthma and atopy in ethnically diverse populations. *Am J Hum Genet* 2000;66:517-26.
- Ober C, Salenko A, Parry R, Cox NJ. A second-generation genomewide screen for asthma-susceptibility alleles in a founder population. *Am J Hum Genet* 2000;67:1154-62.
- Donfack J, Schneider DH, Tan Z, Kurz T, Dubchak I, Frazer KA, et al. Variation in conserved non-coding sequences on chromosome 5q and susceptibility to asthma and atopy. *Respir Res* 2005;6:145-56.
- Kurz T, Hoffjan S, Hayes MG, Schneider D, Nicolae R, Heinzmann A, et al. Fine mapping and positional candidate studies on chromosome 5p13 identify multiple asthma susceptibility loci. *J Allergy Clin Immunol* 2006;118:396-402.
- Weiss LA, Kosova G, Delahanty RJ, Jiang L, Cook EH, Ober C, et al. Variation in ITGB3 is associated with whole-blood serotonin level and autism susceptibility. *Eur J Hum Genet* 2006;14:923-31.
- Weiss LA, Ober C, Cook EH Jr. ITGB3 shows genetic and expression interaction with SLC6A4. *Hum Genet* 2006;120:93-100.
- Lemanske RF Jr. The childhood origins of asthma (COAST) study. *Pediatr Allergy Immunol* 2002;13(suppl 15):38-43.
- Hoffjan S, Nicolae D, Ostrovskaya I, Roberg K, Evans M, Mirel DB, et al. Gene-environment interaction effects on the development of immune responses in the 1st year of life. *Am J Hum Genet* 2005;76:696-704.
- Gern JE, Reardon CL, Hoffjan S, Nicolae D, Li Z, Roberg KA, et al. Effects of dog ownership and genotype on immune development and atopy in infancy. *J Allergy Clin Immunol* 2004;113:307-14.
- Neaville WA, Tisler C, Bhattacharya A, Anklam K, Gilbertson-White S, Hamilton R, et al. Developmental cytokine response profiles and the clinical and immunologic expression of atopy during the first year of life. *J Allergy Clin Immunol* 2003;112:740-6.
- Gern JE, Brooks GD, Meyer P, Chang A, Shen K, Evans MD, et al. Bidirectional interactions between viral respiratory illnesses and cytokine responses in the first year of life. *J Allergy Clin Immunol* 2006;117:72-8.
- Hoffjan S, Ostrovskaya I, Nicolae D, Newman DL, Nicolae R, Gangnon R, et al. Genetic variation in immunoregulatory pathways and atopic phenotypes in infancy. *J Allergy Clin Immunol* 2004;113:511-8.
- Carlson CS, Eberle MA, Rieder MJ, Yi Q, Kruglyak L, Nickerson DA. Selecting a maximally informative set of single-nucleotide polymorphisms for association analyses using linkage disequilibrium. *Am J Hum Genet* 2004;74:106-20.
- Stephens M, Smith NJ, Donnelly P. A new statistical method for haplotype reconstruction from population data. *Am J Hum Genet* 2001;68:978-89.
- Stephens M, Donnelly P. A comparison of bayesian methods for haplotype reconstruction from population genotype data. *Am J Hum Genet* 2003;73:1162-9.
- Lemanske RF Jr, Jackson DJ, Gangnon RE, Evans MD, Li Z, Shult PA, et al. Rhinovirus illnesses during infancy predict subsequent childhood wheezing. *J Allergy Clin Immunol* 2005;116:571-7.
- Villette S, Kyle JA, Brown KM, Pickard K, Milne JS, Nicol F, et al. A novel single nucleotide polymorphism in the 3' untranslated region of human glutathione peroxidase 4 influences lipoxigenase metabolism. *Blood Cells Mol Dis* 2002;29:174-8.
- Goto Y, Yue L, Yokoi A, Nishimura R, Uehara T, Koizumi S, et al. A novel single-nucleotide polymorphism in the 3'-untranslated region of the human dihydrofolate reductase gene with enhanced expression. *Clin Cancer Res* 2001;7:1952-6.
- Huang D, Giscombe R, Zhou Y, Pirskanen R, Lefvert AK. Dinucleotide repeat expansion in the CTLA-4 gene leads to T cell hyper-reactivity via the CD28 pathway in myasthenia gravis. *J Neuroimmunol* 2000;105:69-77.
- Clop A, Marcq F, Takeda H, Pirottin D, Tordoir X, Bibe B, et al. A mutation creating a potential illegitimate microRNA target site in the myostatin gene affects muscularity in sheep. *Nat Genet* 2006;38:813-8.
- Hirota T, Suzuki Y, Hasegawa K, Obara K, Matsuda A, Akahoshi M, et al. Functional haplotypes of IL-12B are associated with childhood atopic asthma. *J Allergy Clin Immunol* 2005;116:789-95.
- Vercelli D, Jabara HH, Arai K, Geha RS. Induction of human IgE synthesis requires interleukin 4 and T/B cell interactions involving the T cell receptor/CD3 complex and MHC class II antigens. *J Exp Med* 1989;169:1295-307.
- Reynolds LE, Conti FJ, Lucas M, Grose R, Robinson S, Stone M, et al. Accelerated re-epithelialization in beta3-integrin-deficient mice is associated with enhanced TGF-beta1 signaling. *Nat Med* 2005;11:167-74.
- Hansen G, McIntire JJ, Yeung VP, Berry G, Thorbecke GJ, Chen L, et al. CD4(+) T helper cells engineered to produce latent TGF-beta1 reverse allergen-induced airway hyperreactivity and inflammation. *J Clin Invest* 2000;105:61-70.
- Nakao A, Sagara H, Setoguchi Y, Okada T, Okumura K, Ogawa H, et al. Expression of Smad7 in bronchial epithelial cells is inversely correlated to basement membrane thickness and airway hyperresponsiveness in patients with asthma. *J Allergy Clin Immunol* 2002;110:873-8.
- Redington AE, Madden J, Frew AJ, Djukanovic R, Roche WR, Holgate ST, et al. Transforming growth factor-beta 1 in asthma. Measurement

- in bronchoalveolar lavage fluid. Am J Respir Crit Care Med 1997;156:642-7.
42. Groneberg DA, Witt H, Adcock IM, Hansen G, Springer J. Smads as intracellular mediators of airway inflammation. Exp Lung Res 2004;30:223-50.
43. Eder W, Klimecki W, Yu L, von Mutius E, Riedler J, Braun-Fahrlander C, et al. Opposite effects of CD 14/-260 on serum IgE levels in children raised in different environments. J Allergy Clin Immunol 2005;116:601-7.
44. Williams LK, McPhee RA, Ownby DR, Peterson EL, James M, Zoratti EM, et al. Gene-environment interactions with CD14 C-260T and their relationship to total serum IgE levels in adults. J Allergy Clin Immunol 2006;118:851-7.
45. Zambelli-Weiner A, Ehrlich E, Stockton ML, Grant AV, Zhang S, Levett PN, et al. Evaluation of the CD14/-260 polymorphism and house dust endotoxin exposure in the Barbados Asthma Genetics Study. J Allergy Clin Immunol 2005;115:1203-9.
46. Bazan-Socha S, Bukiej A, Pulka G, Marcinkiewicz C, Musial J. Increased expression of collagen receptors: alpha1beta1 and alpha2beta1 integrins on blood eosinophils in bronchial asthma. Clin Exp Allergy 2006;36:1184-91.
47. Nguyen TT, Ward JP, Hirst SJ. beta1-Integrins mediate enhancement of airway smooth muscle proliferation by collagen and fibronectin. Am J Respir Crit Care Med 2005;171:217-23.

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