

# Dense mapping of chromosome 12q13.12-q23.3 and linkage to asthma and atopy

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**Background:** Asthma is a complex disease characterized by a high prevalence of allergic diathesis and the almost ubiquitous presence of upper airway disease (eg, rhinitis). Previously, we observed linkage of asthma among Afro-Caribbean families to markers in chromosome 12q, which contains a number of genes encoding for products closely related to allergic airway inflammation and disease.

**Objective:** To identify susceptibility loci in chromosome 12q contributing to the genetics of upper and lower airway diseases and to expand the region to include genes encoding IFN- $\gamma$  (*IFNG*) and one of the signal transducers and activators of transcription (*STAT6*), we conducted further linkage studies among 33 multiplex families.

**Methods:** We characterized 528 subjects from Barbados for asthma; 82% were characterized for allergic rhinitis. Two-point and multipoint linkage analysis of 22 microsatellite markers (spanning ~79 centimorgan) was performed.

**Results:** Affected sib-pair analysis revealed significant evidence for linkage to asthma over approximately 30 cM ( $P < .05$  to  $.002$ ), with the best evidence for linkage at a CA repeat polymorphism in the first intron of *IFNG* in 12q21.1 ( $P = .002$ ). Evidence of linkage to allergic rhinitis was observed in the same region (*D12S313*,  $P = 0.006$ , and *IFNGCA*,  $P = .01$ , respectively). Multipoint linkage analysis also provided evidence for linkage to asthma, with the best nonparametric link-

age analysis score at *D12S326* (nonparametric linkage score = 3.8,  $P = .0008$ ). Modest evidence for linkage to allergic rhinitis was observed next to *D12S326* at *D12S1052* ( $P = .036$ ).

**Conclusions:** Our findings suggest that (1) one or more loci in the chromosome 12q13.12–q23.3 region are contributing to the expression of the clinical phenotype asthma and the strongest evidence for linkage is in a region near the gene encoding *IFNG* and (2) a susceptibility locus for both asthma and allergic rhinitis maps to this region. (*J Allergy Clin Immunol* 1999;104:485–91.)

**Key words:** Asthma, allergic rhinitis, linkage analysis, chromosome 12q, IFN- $\gamma$

Asthma is a complex disease in which both genetic and environmental factors contribute to the overall phenotype. By using both the candidate gene and genome-wide screening approaches, multiple loci throughout the human genome have been implicated in the etiology of asthma and asthma-associated phenotypes.<sup>1–3</sup> Previously, our group and others provided strong evidence for linkage to asthma and atopy to a rather large region (spanning >35 cM) on human chromosome 12q15–q24.1.<sup>3–6</sup> This region contains a number of candidate genes important for asthma and atopy, including *IFNG*, stem cell factor (*SCF*), leukotriene A4 hydrolase (*LTA4H*), insulin-like growth factor-1 (*IGF1*), the  $\beta$ -subunit of nuclear factor- $\kappa$ B (*NFYB*), and B-cell translocation gene 1 (*BTG1*). In recent years the study of how cytokines regulate gene transcription has led to the characterization of signal transducers and activators of transcription (STATs). Of these, STAT6 is of particular interest in the study of atopy because it has been shown to be essential for both IL-4 signaling and T<sub>H2</sub> development<sup>8,9</sup> and IL-13.<sup>10,11</sup> Svaren et al<sup>12</sup> reported that *STAT6* is clustered with *Nab2*, which maps 0.6 cM downstream from *Gli*, a gene that has been mapped to chromosome 12q13.2. Undoubtedly there are a host of other candidate genes within chromosome 12q that may be contributing to the pathogenesis of asthma and atopy but that to date have not been identified.

Most of the genetic studies of asthma and atopy have focused primarily on asthma as a discrete trait or on phenotypes associated with asthma (eg, bronchial hyperresponsiveness and wheezing) and/or phenotypes associated with IgE (eg, total IgE, skin test sensitivity, and atopy). These studies have not examined linkage to allergic rhinitis despite the proposed notion that asthma and

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<sup>†</sup>Dr David G. Marsh was actively involved in the design and implementation of this study. He passed away on March 29, 1998, after a heroic battle with cancer. This paper is dedicated in his memory.

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**Abbreviations used**

ASP: Affected sib-pair  
 IBD: Identity by descent  
 NPL: Nonparametric linkage  
 RLU: Relative light unit  
 TDT: Transmission disequilibrium test

allergic rhinitis may be "a continuum of inflammation within one common airway."<sup>13</sup> Asthma and allergic rhinitis are diseases that are both characterized by airway inflammation and mediated by hypersensitivity and other common immunologic mechanisms. Upper and lower airway inflammation involves a similar array of cellular mechanisms and inflammatory mediators, including mast cells, eosinophils, macrophages, and T lymphocytes.<sup>14-20</sup> There has been a tremendous focus on the overlapping roles of leukotrienes and histamines as critical inflammatory mediators in both the upper and lower airways.<sup>21-27</sup> A second common denominator of these 2 clinical phenotypes is their strong association with allergy, a determinant of the inflammatory process. For example, despite the evidence suggesting a distinction between nonatopic and atopic asthma,<sup>28,29</sup> both are characterized by increased eosinophil numbers in the blood, sputum, and bronchial alveolar lavage fluid, bronchial mucosa infiltration with cells expressing IL-4 and IL-5, and elevated numbers of submucosal cells expressing messenger (m)RNA encoding RANTES, monocyte chemoattractant protein-3, GM-CSF, IL-5, and IL-3.<sup>30,31</sup>

In our initial study 10 microsatellite markers on chromosome 12q were analyzed in 29 multiplex Afro-Caribbean families. Evidence of linkage to asthma was strongest between *D12S379* and *D12S360* ( $P = .018$  to  $.001$ ) by 2-point affected sib-pair (ASP) and multipoint affected relative analysis.<sup>4</sup> To pursue the hypothesis that loci in this region contribute to the expression of phenotypes associated with inflammation and hyperresponsiveness of the human airways, we continued with our genetic studies in Afro-Caribbean multiplex families from Barbados by using 2-point, ASP, and multipoint analysis and a denser map of polymorphic markers, with a focus on the binary traits of asthma and allergic rhinitis. Given the potential importance of *IFNG* and *STAT6* in the pathophysiology of asthma and atopy, we typed additional markers in these loci, including a marker of a CA repeat polymorphism in the first intron of the *IFNG* gene (*INFG-CA*) and 2 flanking markers (*D12S335*, *D12S313*) and 2 markers flanking the *STAT6* gene (*D12S390*, *D12S398*).

**METHODS****Study subjects**

Of the 528 subjects from Barbados presented here, 29 kindreds (507 subjects) have been previously described.<sup>4</sup> An additional 21 members in 4 new kindreds were recruited for this study. All families were selected on the basis of an asthma proband. There were a total of 527 Afro-Caribbean participants and 1 Caucasian subject (the father of a proband), the total of which included 154 asthmatic subjects (50% male) and their family members. Information regarding

allergic rhinitis was available for 82% of these individuals; 168 of 432 (39%) subjects (42% male) met the criteria for allergic rhinitis. Among asthmatic subjects who provided data on their allergic rhinitis status, 99 of 154 (70%) also had allergic rhinitis, and 99 of 168 (59%) subjects with allergic rhinitis had asthma in this sample. The age- and sex-adjusted geometric mean total IgE concentration for the population was 482 ng/mL (2.68 log ng/mL); the geometric mean concentrations for subjects with asthma and allergic rhinitis were 1072 ng/mL (3.03 log ng/mL) and 1622 ng/mL (3.21 log ng/mL), respectively. All participants gave their informed consent as approved by The Johns Hopkins University Institutional Review Board.

**Phenotypes**

**Asthma.** Asthma was defined as both (1) a reported history of asthma determined by using a standardized questionnaire adapted from the ongoing National Heart, Lung, and Blood Institute *Collaborative Study on the Genetics of Asthma*,<sup>32</sup> which includes information on respiratory symptoms (cough, wheeze, shortness of breath, chest tightness) and a history of physician-diagnosed asthma (past/current), and (2) confirmation of asthma by an interview of the patient with our clinician (RPN), one of the primary diagnosticians of asthma in Barbados.

**Allergic rhinitis.** Allergic rhinitis was defined as both (1) a positive answer to the questions "Have you ever had hay fever or a runny or stuffy nose accompanied by sneezing and itching when you did not have a cold or flu?" and "Have you ever had a runny or stuffy nose with exposure to any of these circumstances (pollens, house dust, mold, moist or damp places, cats, dogs, other animals or birds, weather changes, chemicals, drugs, anything else)?" and (2) positive IgE antibody concentration(s) to at least one of the following: specific IgE antibodies to a multiallergen composite and/or specific IgE antibodies to the mite *Blomia tropicalis* (Blo t M). Individuals reporting an uncertain history of allergic rhinitis (6 subjects) were categorized as being of unknown status.

**IgE measurements.** Total IgE concentrations were measured as previously described<sup>4,33</sup> by using the chemiluminometric Magic Lite immunoassay (Magic Lite; ALK, Copenhagen, Denmark/CIBA-Corning, Medfield, Mass) and were adjusted for all individuals for both age and sex.

Specific IgE antibodies were measured by using the multiallergen analysis (Magic Lite), which is a proprietary 2-site chemiluminescence assay using paramagnetic particles to which standardized quality extracts are covalently coupled by optimized procedures. The multicomponent system contains a mixture of allergen-coupled particles of 20 different aeroallergens (molds, weed pollens, tree pollens, grass pollens, animal dander, cockroach, and house dust mites) and has been described elsewhere.<sup>33</sup> The IgE antibody levels in the patients' samples are expressed in terms of the arbitrary units per milliliter that have been assigned to the standard serum, and values were adjusted for all individuals for both age and sex. Multiallergen screening tests have been shown to be highly efficient in identifying subjects with atopy, and the sensitivity and specificity of the Magic Lite system has been shown to be reliable and is widely used.<sup>34-36</sup>

Additional specific IgE antibody levels were tested by using separate solid-phase extracts of *B tropicalis* (Blo t M), one of the major mite species and arguably the most important allergen in Barbados. Each sample was assayed in duplicate, and values differing by 10% or greater were repeated until the coefficient of variation was within 10%. The IgE antibody concentrations were expressed in terms of arbitrary units per milliliter that were internally consistent. IgE antibody measurements were log-transformed to minimize skewness. For individuals with Blo t M IgE antibody titers above the lowest detectable value (1.0 U/mL), titers were adjusted for age (20 years) by linear regression analysis (there was no difference according to sex).

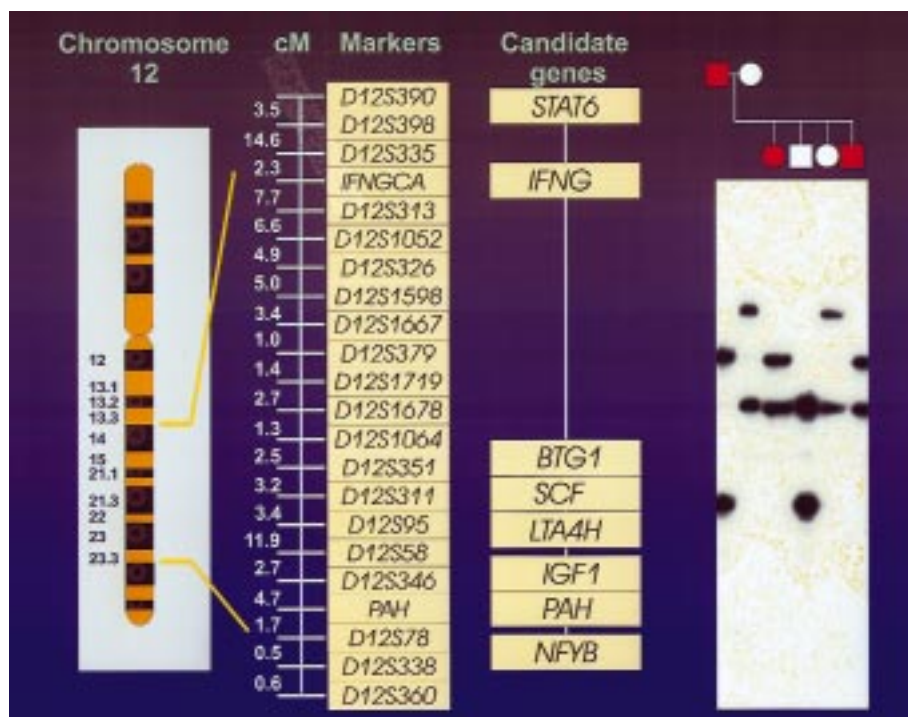


**FIG 1.** Linkage map of human chromosome 12q13.12-q23.3 based on a sex-averaged linkage map of the 22 markers analyzed with the CRIMAP 2.4 Kosambi mapping function.<sup>38</sup> The map includes the following genes: *STAT6*, *IFNG*, B-cell translocation gene 1 (*BTG1*), stem cell factor (*SCF*), leukotriene A(4) hydrolase (*LTA4H*), insulin-like growth factor-1 (*IGF1*), phenylalanine hydroxylase (*PAH*); and the  $\beta$ -subunit of nuclear factor-Y (*NFYB*). Physical distances of the genes in relation to the markers are approximate based on data obtained from <http://cedar.genetics.soton.ac.uk>.

We defined a threshold in the subjects from Barbados between the low and high phenotypes for specific IgE antibodies as the concentration that minimized the number of high total IgE responders with atypically low specific IgE antibodies and the number of low total IgE responders with atypically positive high IgE antibodies. After plotting the cumulative distribution frequencies for log-specific IgE antibody in the low and high total IgE subsets (as the cumulative percentage of the total number of subjects in each subgroup), the difference curve for these 2 sets of data was calculated. The maximum of this difference curve gives the best estimate for a threshold for specific IgE antibody. For multiallergen IgE antibody, which was normally distributed, this maximum difference was 0.4 log U/mL. Because of a bimodal distribution of Blo t M IgE antibody among low responders (ie, responders vs nonresponders), we selected the optimal cut-off point (0.6 log U/mL) among responders only by using the Kernel density estimation (SYSTAT<sup>37</sup>).

## Genotyping

Twenty-two markers on chromosome 12q were analyzed by using the methodology previously described.<sup>4</sup> We used the same set of 10 microsatellite markers from our previous study plus 12 additional markers: *D12S390*, *D12S398*, *D12S335*, *D12S313*, *IFNGCA*, *D12S326*, *D12S1667*, *D12S1719*, *D12S1678*, *D12S1598*, *D12S58*, and *D12S346* to examine this region more closely.

A sex-averaged linkage map of the markers analyzed was prepared by using the Kosambi mapping function with CRIMAP 2.4.<sup>38</sup> The best supported order obtained from our data was compared with the physical map of Krauter et al<sup>7</sup> and other published maps (eg, <http://cedar.genetics.soton.ac.uk>). Marker distances and orders varied slightly from our previous report.<sup>4</sup> The total chromosomal region covered by these 22 markers spanned approximately 79 cM.

## Statistical analyses

ASP linkage analyses were performed by using the program SIBPAL included in the SAGE 3.1 package.<sup>39</sup> Multipoint linkage analyses were performed by using the program GENEHUNTER.<sup>40</sup> The nonparametric linkage (NPL) score represents identity-by-descent (IBD) sharing among all affected pedigree members. Extended pedigrees occasionally had to be trimmed to meet computational constraints, and in these circumstances some informative individuals (ie, subjects with genotypic information and affected individuals) were deleted.

Because of previous studies showing evidence of association between a CA repeat polymorphism of the gene encoding *IFNG* and a higher production of IFN- $\gamma$ ,<sup>41</sup> we conducted the transmission disequilibrium test (TDT) to further examine for evidence of linkage disequilibrium with the CA repeat and the unobserved gene regulation of asthma and allergic rhinitis in this population. TDTs were performed by using the program by DLD.<sup>42</sup>

## RESULTS

### Sib-pair analysis

Four families were added to the original dataset and included in these analyses, yielding a total of 528 subjects from Barbados (33 multiplex kindreds). The 10 markers that were described previously<sup>4</sup> plus 12 additional markers in this region were analyzed by using ASP linkage analysis (Fig 1). Table I illustrates the findings from ASP analysis of each marker individually. Markers between *D12S313* and *D12S95* (spanning ~30 cM) showed significant evidence of linkage demonstrated by

**TABLE I.** ASP analyses of chromosome 12q markers for asthma and allergic rhinitis in 33 Barbados kindreds

| Locus    | Asthma*                                                 |                       | Allergic rhinitis*                                      |                       |
|----------|---------------------------------------------------------|-----------------------|---------------------------------------------------------|-----------------------|
|          | Percentage alleles shared (full/half sibs) <sup>†</sup> | P values <sup>‡</sup> | Percentage alleles shared (full/half sibs) <sup>†</sup> | P values <sup>‡</sup> |
| D12S390  | 50/28                                                   | —                     | 53/25                                                   | —                     |
| D12S398  | 55/28                                                   | .07                   | 57/18                                                   | —                     |
| D12S335  | 58/26                                                   | .07                   | 55/24                                                   | —                     |
| IFNGCA   | 61/31                                                   | <b>.002</b>           | 60/27                                                   | <b>.01</b>            |
| D12S313  | 60/28                                                   | <b>.008</b>           | 56/33                                                   | <b>.006</b>           |
| D12S1052 | 56/23                                                   | —                     | 55/27                                                   | .09                   |
| D12S326  | 63/28                                                   | <b>.009</b>           | 54/26                                                   | —                     |
| D12S1598 | 61/26                                                   | <b>.02</b>            | 49/30                                                   | —                     |
| D12S1667 | 56/26                                                   | —                     | 50/25                                                   | —                     |
| D12S379  | 62/27                                                   | <b>.003</b>           | 51/25                                                   | —                     |
| D12S1719 | 55/21                                                   | —                     | 54/22                                                   | —                     |
| D12S1678 | 59/22                                                   | —                     | 54/24                                                   | —                     |
| D12S1064 | 60/21                                                   | <b>.04</b>            | 56/24                                                   | —                     |
| D12S351  | 61/23                                                   | .08                   | 52/25                                                   | —                     |
| D12S311  | 60/30                                                   | <b>.007</b>           | 53/28                                                   | —                     |
| D12S95   | 61/28                                                   | <b>.01</b>            | 53/27                                                   | —                     |
| D12S58   | 56/25                                                   | —                     | 53/28                                                   | —                     |
| D12S346  | 53/26                                                   | —                     | 51/28                                                   | —                     |
| PAH      | 59/24                                                   | .07                   | 55/23                                                   | —                     |
| D12S78   | 57/25                                                   | —                     | 56/26                                                   | —                     |
| D12S338  | 56/27                                                   | —                     | 52/21                                                   | —                     |
| D12S360  | 59/25                                                   | <b>.05</b>            | 58/25                                                   | —                     |

\*There were a total of 345 full and 219 half sib-pairs in the Barbados families. The number of asthmatic full/half sib-pairs was 63/30. The number of allergic rhinitis full/half sib-pairs ranged from 58/33.

<sup>†</sup>Represents the mean proportion of marker alleles shared IBD.

<sup>‡</sup>P values of .05 or greater are shown in bold; P values of .10 or greater are not presented.

increased allele-sharing in concordant asthmatic sib-pairs ( $P < .05$  to  $.002$ ), with the best evidence for linkage at marker *IFNGCA* ( $P = .002$ ). Significant evidence of linkage to allergic rhinitis was observed at *D12S313* and *IFNGCA* ( $P = .006$  and  $P = .01$ , respectively); the significance of this evidence at *D12S313* increased when asthmatic subjects were removed from the allergic rhinitis dataset ( $P = .0006$ , data not shown), although there were only 14 full- and 8 half sib-pairs in this latter analysis.

### Multipoint linkage mapping

These marker data were further analyzed by using the multipoint linkage program GENEHUNTER<sup>40</sup> (Fig 2). Maximal evidence for linkage to asthma occurred at *D12S326* (NPL = 3.8,  $P = .0008$ ), but significant NPL scores ranged from *D12S335* to *D12S95* (2.6 to 1.7, respectively; ~33 cM). A modest NPL score was evident for allergic rhinitis between *IFNGCA* and *D12S1052* (1.3 to 1.8), with the best peak at *D12S1052* (NPL = 1.8,  $P = .036$ ). The average information content of the map based on 22 markers was 81.3% for asthma and 80.0% for allergic rhinitis.

### Test for linkage disequilibrium of the CA repeat in the first intron of *IFNG*

A total of 8 different alleles were observed for *IFNG*-CA in the Barbados dataset; alleles 104 and 106 were observed at frequencies of 31% and 40%, respectively. By using the TDT, there was no evidence for linkage at the .05 level of significance with either asthma or allergic rhinitis (data not shown).

### DISCUSSION

In this study we investigated the positioning of susceptibility loci responsible for asthma and allergic rhinitis in human chromosome 12q13.12–q23.3 (spanning a region of ~79 cM). Because of our initial findings of linkage to asthma and *D12S379*,<sup>4</sup> we conducted further analyses in this 12q21.33 region with additional markers and also typing markers upstream of *D12S379*, including markers flanking and including *IFNG* (12q21.1) and *STAT6* (12q13.12–q13.13). Additional information derived from analyzing more families in Barbados, denser mapping in the region of interest, and the application of multipoint linkage methods support our previous hypothesis that one or more genes controlling the expression of the clinical phenotype of asthma are located in this general region on chromosome 12q.

Although we did not analyze markers upstream from *D12S379* (including *IFNG*) in our original study of Barbados subjects,<sup>4</sup> we did conduct marker analysis for *IFNG* (but not *IFNGCA*) in an inbred white population (Amish) for the log[total IgE] phenotype (asthma data were unavailable) and found no evidence for linkage. In the current study our best evidence for linkage for both asthma and allergic rhinitis using ASP analysis was for markers at or near (within 6.5 cM; distance according to <http://cedar.genetics.soton.ac.uk>) the gene *IFNG* (Table I), a cytokine important in regulating immune responses. There was no evidence for linkage to the *STAT6* markers (*D12S390*, *D12S398*) for asthma or allergic rhinitis by 2-point or multipoint analyses. Multipoint linkage analysis suggested a maximum NPL score for asthma and allergic rhinitis of approximately 5 to 12 cM downstream at *D12S326* and *D12S1052*, respectively (Fig 2), suggesting that the susceptibility locus is not *IFNG* but a nearby unidentified gene. Significant NPL scores were observed for asthma over a very large region (~40 cM), which is suggestive of multiple susceptibility loci for asthma, but not necessarily allergic rhinitis, within this region. However, linkage analysis depends on informative meioses and an effective sample size for analysis, and therefore we may observe a considerable range in the confidence limits of the actual susceptibility locus in our particular set of multiplex families and microsatellite markers.

Our data suggest that a susceptibility locus for allergic rhinitis either overlaps with the putative asthma locus or lies in close proximity to it. A common feature of the 2 entities, asthma and allergic rhinitis, is of course the allergic diathesis. Evidence for linkage to high total IgE

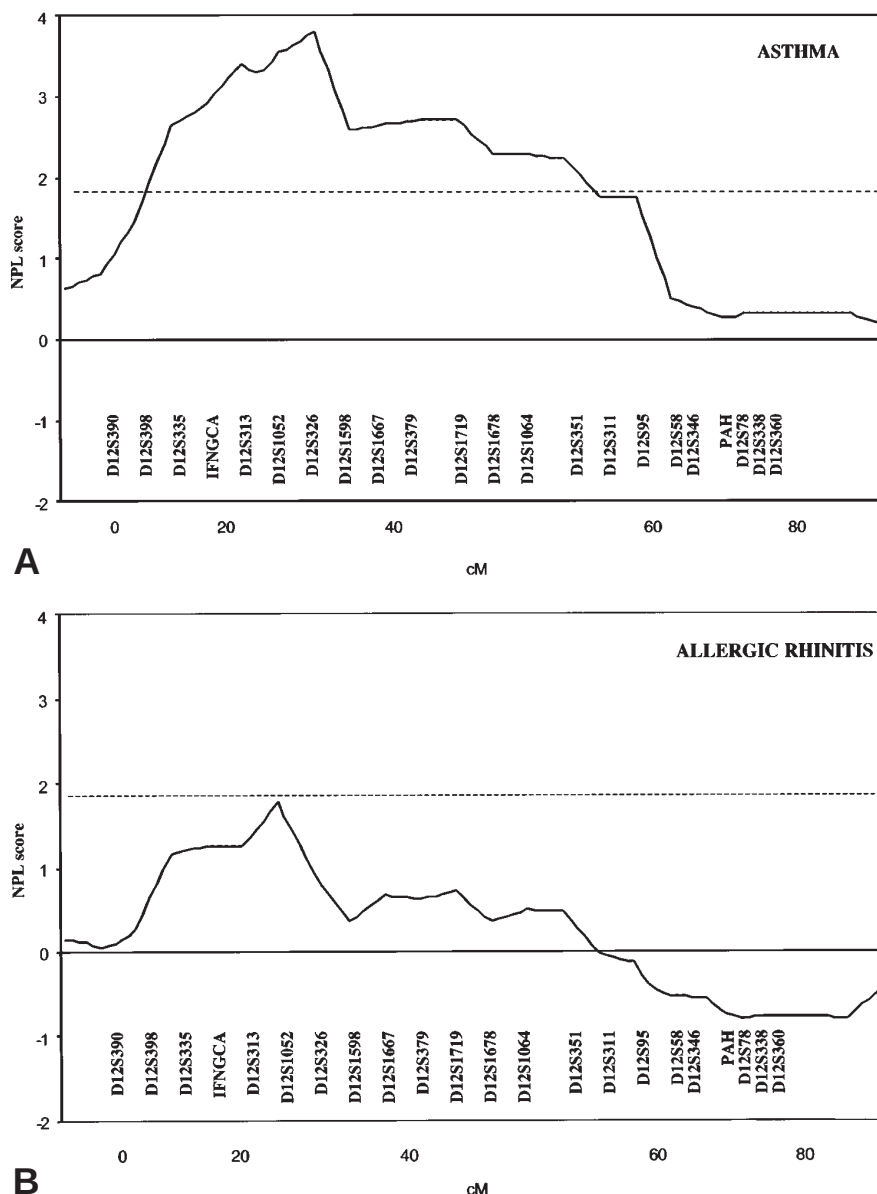


**FIG 2.** Multipoint IBD affected relative analysis of chromosome 12q markers in 33 Barbadian kindreds for asthma (A) and allergic rhinitis (B). The x axis illustrates the map positions of the markers by using the Kosambi mapping function for both CRIMAP 2.4<sup>38</sup> and GENEHUNTER (Version 1.0;<sup>40</sup> under the "fixed" option and the locus order indicated by the physical map of Krauter et al<sup>7</sup>), and the y axis is the NPL score. The horizontal, small-hatched line indicates level of significance ( $P < .05$ ).

has previously been observed within this large region on chromosome 12q for the families from Barbados and 2 distinct Caucasian populations,<sup>4,6</sup> and it is possible that the putative locus for which we observe linkage to both asthma and allergic rhinitis is actually an atopy locus.

In the current study we analyzed nonasthmatic subjects with allergic rhinitis to determine whether evidence for linkage in this region to allergic rhinitis was being solely driven by the asthmatic subjects in the group. This approach did not diminish evidence for linkage to an unobserved gene controlling risk for allergic rhinitis but increased the level of significance (D12S313,  $P = .0006$ ).

Unfortunately, adjusting allergic rhinitis for asthma status severely reduced the number of affected full/half sibs (to only 14 full sibs, 8 half sibs), and therefore a more definitive conclusion must await the collection of nonasthmatic subjects with allergic rhinitis for future analyses. Although it can be argued that a certain degree of bias is built into this study because all families were selected for asthma, we argue that given the high degree of coexistence of asthma and allergic rhinitis, it would be most difficult to recruit families for atopic asthma for whom no members had allergic rhinitis and, conversely, families for allergic rhinitis for whom no members had

atopic asthma. Our criteria for identifying allergic rhinitis is based on the patient's history and the presence of clinically relevant specific immunoresponsiveness (eg, serum IgE antibody concentrations). Although objective widely acceptable measures of upper airway function that distinguish allergic rhinitis from the healthy state have not been developed, the temporal relationship between symptoms and allergic sensitization to the most common aeroallergens is so strong that the definition of the allergic rhinitis phenotype should be considered very solid.

IFN- $\gamma$  is unique from IFN- $\alpha$  and IFN- $\beta$  because it has 3 introns; the other 2 have no introns.<sup>43</sup> A recent study suggested that a CA repeat polymorphism in the first intron of the *IFNG* gene is associated with a higher production of IFN- $\gamma$ .<sup>41</sup> Previously, it was shown that the CA repeat polymorphism was associated with insulin-dependent diabetes mellitus among Japanese patients,<sup>44</sup> but Pociot et al<sup>45</sup> were unable to replicate these findings among insulin-dependent diabetes mellitus multiplex families from Finland. More recently, Hayden et al<sup>46</sup> reported that they found no polymorphisms in the *IFNG* gene that were associated with atopic asthma; however, the group screened for mutations in the exons and 5' flanking region only. In the Barbados study we found no evidence for overall transmission disequilibrium of allele frequencies among asthmatic subjects by the TDT, despite evidence of linkage by 2-point ASP and multipoint NPL analyses. Because the TDT was originally designed as a test for linkage in the presence of association,<sup>47</sup> it is not surprising that the approach is less powerful in the absence of association. We saw no evidence of allelic association between the CA repeat and asthma or rhinitis in this population. Unless a trait locus causes a major effect, modest numbers of families may not yield sufficient power to detect linkage by using the TDT alone. Using data from the 33 families from Barbados and the program SLINK,<sup>48</sup> we simulated a 10-allele marker absolutely linked to a susceptibility locus for asthma but could detect no linkage disequilibrium (data not shown). Lastly, because the CA repeat is in the intron and is therefore most likely nonfunctional, it is still possible that a functional mutation elsewhere in the *IFNG* gene causes susceptibility to asthma and rhinitis. If so, this functional mutation appears not to be in linkage disequilibrium with the CA repeat marker. Further studies are necessary to elucidate this question.

Our study is based on data from families of asthmatic subjects in Barbados, a population in which exposure to common indoor allergens, especially the storage mite *B tropicalis*, is pervasive, and therefore expression of atopic disease is pronounced. Previously, we have described the profound impact that chronic exposure to high levels of indoor allergens apparently has on residents of Barbados,<sup>4</sup> which is most evident in the very high mean total IgE concentration for the total study population (482 ng/mL), especially among atopic subjects (mean >1000 ng/mL). Moreover, serum IgE does not appear to be driven by parasitic infection in this population; there is in fact a notable absence of extracellular

parasitic disease in Barbados (Levett, 1998, unpublished data), which is typically endemic in tropical regions.

Recent advances in the studies of inflammatory mediators involved in the pathophysiology of asthma and allergic rhinitis have weakened the conventional notion that these 2 atopic disorders are separate entities and have contributed to the belief that asthma and allergic rhinitis are essentially the same inflammatory disease of human airways.<sup>13</sup> The strongest evidence in support of this hypothesis is the high rate of co-occurrence of these two entities.<sup>49-53</sup> It has been reported that between 60% and 80% of subjects with allergic asthma also have rhinitis<sup>49,52</sup>; however, a recent study by Togias' group<sup>53</sup> with a highly sensitive standardized tool determined that 98.9% of 473 atopic asthmatic subjects surveyed had allergic rhinitis. Conversely, allergic rhinitis is regarded as a risk factor for asthma because 20% to 38% of subjects with allergic rhinitis also have asthma.<sup>49,52</sup> We contend that a number of overlapping genes are important for both asthma and allergic rhinitis. Given the epidemiologic data, it can be argued that these 2 phenotypes might represent a continuum of total airway disease, whereby those with less severe disease will express rhinitis without asthma, and those with more severe disease express both phenotypes. An alternative hypothesis is that there are common genes associated with both asthma and allergic rhinitis, with additional genes determining the ultimate risk for asthma.

In summary, both the ASP analyses and multipoint affected-relative analyses in this enlarged set of Afro-Caribbean subjects suggest that one or more loci within the human chromosome 12q13.12-q23.3 region control asthma and that possibly a susceptibility locus for both asthma and allergic rhinitis is in this region as well. Confirmation of our finding would support the recent argument that these 2 diseases are the same entity and warrants further study. Finer mapping in this region is required to determine the most likely candidate genes. Replication of linkage to these traits in other populations is also necessary to determine the relevance of our findings.

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