Letter to the Editor

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Novel Vitiligo Susceptibility Loci on Chromosomes 7 (*AIS2*) and 8 (*AIS3*), Confirmation of *SLEV1* on Chromosome 17, and Their Roles in an Autoimmune Diathesis

To the Editor:

Generalized vitiligo (MIM 193200) is a common acquired disorder in which patches of white skin and overlying hair result from loss of pigment-forming melanocytes (reviewed in Bolognia et al. [1998], Kovacs [1998], and Hann and Nordlund [2000]), apparently because of a noninflammatory, T cell autoimmune response (reviewed in Ongenae et al. [2003]). Vitiligo occurs in 0.38% of whites (Howitz et al. 1977) and, in 23% of cases, is associated with other autoimmune disorders, particularly including autoimmune thyroid disease, pernicious anemia, systemic lupus erythematosus, Addison disease (Alkhateeb et al. 2003), and adult-onset insulin-dependent diabetes mellitus (our unpublished data). This complex of associated multiple autoimmune diseases most likely results from combinations of genes, some predisposing to an inherited autoimmune diathesis and others to specific forms of autoimmune disease. In the past, the association of various multiple autoimmune diseases has been termed "autoimmune polyendocrine syndrome, type 2" (APS2 [MIM 269200]), with a number of descriptive subcategories, the biological bases for which are uncertain (reviewed in Betterle et al. [2002]).

Vitiligo is a polygenic, multifactorial disorder (Majumder et al. 1988; Nath et al. 1994; Arcos-Burgos et al. 2002; Alkhateeb et al. 2003), with frequent family clustering (Mehta et al. 1973; Carnevale et al. 1980; Goudie et al. 1983; Hafez et al. 1983; Das et al. 1985; Majumder et al. 1993; Alkhateeb et al. 2003), and ~20% of probands having at least one affected first-degree relative (Alkhateeb et al. 2003). We previously described a genome scan of 71 white multiplex families with vitiligo, and we mapped *AIS1* (autoimmune susceptibility 1), a locus in chromosome segment 1p31.3-p32.2 that apparently confers susceptibility to generalized vitiligo and associated autoimmune disorders, as well as seven additional suggestive linkage signals on chromosomes 1, 7, 8, 11, 19, and 22 (Alkhateeb et al. 2002; Fain et al. 2003).

We have now extended this study to a total of 102 white multiplex families with vitiligo, providing substantially increased power for genomewide linkage analysis. Families were ascertained principally from the Vitiligo Society (U.K.) and the National Vitiligo Society (U.S.A.), as described elsewhere (Fain et al. 2003). Phenotypes were checked carefully by history, lesion maps, and, in most cases, physical examination and/or photographs; individuals for whom the phenotype was at all questionable were excluded. DNA was prepared from peripheral blood by standard methods, and genotyping was performed on a total of 660 individuals (300 affected with vitiligo; 192 females, 108 males) for 382 autosomal microsatellite markers from the ABI Prism 10-cM Linkage Mapping Set LMSv2-MD10, with manual checking of all genotypes by at least two people to minimize data errors, as described elsewhere (Fain et al. 2003). Multipoint nonparametric linkage analyses were performed using Allegro (Gudbjartsson et al. 2000). Heterogeneity testing between autoimmune and nonautoimmune families was performed using a predivided sample test (Morton 1956; Ott 1999).

As shown in table 1, analysis of the extended 102-family cohort provides continued strong support for *AIS1* at 73.7 cM on chromosome 1p (LOD = 5.59; P = .000000279) (all positions have been updated in accordance with the deCODE genome map [Kong et al. 2002]). In addition, two other signals that previously were only suggestive now achieve threshold criteria for significant linkage (Lander and Kruglyak 1995). These loci, now designated "*AIS2*" at 89.4 cM on chromosome 7 (LOD = 3.73; P = .0000208) and *AIS3* at 54.2 cM on chromosome 8 (LOD = 3.36; P = .0000418), thus represent candidates for confirmation by analysis of a replicate family cohort. Our data also provide support for a locus at 4.3 cM on chromosome 17 (LOD = 3.07; P = .0000852), most likely corresponding to *SLEV1,* a locus detected in multiplex families with lupus that also segregate cases of vitiligo (Nath et al. 2001). Three linkage signals that were suggestive in our previous analysis of 71 families (Fain et al. 2003)—at 12.2 cM on chromosome 1p, 4.1 cM on 11p, and 107.8 cM on 19q—fell below the threshold for suggestive linkage at the 102-family level. However, we detected two new linkage

Table 1

Suggestive and Significant LODs (and *P* Values) for 71 and 102 Multiplex White Families with Vitiligo

Chromosome and Distance	Marker(s)	LOD (*P*) in 71 Families[a]	LOD (*P*) in 102 Families
1:			
12.2 cM	D1S214	2.17 (.000335)	NS
73.7 cM[b]	D1S2797-D1S2890	***5.56 (.000000282)***	***5.59 (.000000279)***
7:			
89.4 cM[c]	D7S669	2.87 (.000131)	***3.73 (.0000208)***
8:			
54.2 cM[d]	D8S505	1.95 (.00135)	***3.36 (.0000418)***
9:			
88.1 cM	D9S167-D9S283	NS	2.34 (.000238)
11:			
4.1 cM	D11S4046-D11S1338	1.93 (.00142)	NS
13:			
109.4 cM	D13S173	NS	2.30 (.000563)
17:			
4.3 cM[e]	D17S849-D17S831	NS	***3.07 (.0000852)***
19:			
34.9 cM	D19S221-D19S226	2.45 (.000388)	2.62 (.000254)
107.8 cM	D19S210	2.31 (.000551)	NS
22:			
7.7 cM	D22S420-D22S539	2.30 (.000561)	2.98 (.000106)

Note.—Only linkage signals achieving significant (LOD $\geq$ 3.3 and/or $P \leq .000049$ [shown in boldface italics]) or suggestive (LOD $\geq$ 1.86 and/or $P \leq .0017$) de novo linkage thresholds (Terwilliger and Ott 1994; Lander and Kruglyak 1995; Nyholt 2000) are reported; the significant linkage threshold for independent replication (i.e., *SLEV1*) is $P \leq .01$ (Lander and Kruglyak 1995). NS denotes not suggestive. Genome positions have been updated using the deCODE genome map (Kong et al. 2002) and sequence-based STS map (National Center for Biotechnology Information). All gene symbols have been approved by the HUGO gene nomenclature committee.

[a] Fain et al. 2003.
[b] *AIS1*.
[c] *AIS2*.
[d] *AIS3*.
[e] *SLEV1*.

signals that met criteria for suggestive linkage (Lander and Kruglyak 1995)—at 88.1 cM on chromosome 9q (LOD = 2.34; $P = .000238$) and at 109.4 cM on 13q (LOD = 2.30; $P = .000563$)—that represent candidates for follow-up extension and replication linkage studies.

The 102 study families were selected solely on the basis of having multiplex cases of vitiligo. Nevertheless, half of the families segregated only vitiligo, whereas the other half also segregated various others of the vitiligo-associated autoimmune diseases (autoimmune thyroid disease, pernicious anemia, lupus, Addison disease, or adult-onset autoimmune diabetes mellitus) (Alkhateeb et al. 2002) (table 2). This provided an obvious basis for phenotypic stratification of the 102 families into autoimmunity-associated versus nonautoimmunity-associated family subgroups and analysis of the four a priori significant linkage signals in each subgroup. As shown in table 3, the *AIS1*, *AIS2*, and *SLEV1* linkage signals derive principally from the autoimmunity-associated family subgroup and thus may predispose to a vitiligo-associated autoimmunity diathesis. Indeed, the *AIS1* and *SLEV1* LODs increased substantially on family stratification, even though the number of families was reduced by half. *AIS1* was originally mapped in a large family with vitiligo, Hashimoto disease, and 21-hydroxylase autoantibody positivity (a preclinical marker for Addison disease) (Alkhateeb et al. 2002), and *SLEV1* was originally mapped in families segregating lu-

Table 2

Vitiligo-Associated Autoimmune Diseases in the 51 Autoimmunity-Associated Families

Autoimmune Disease	No. of Affected Families	No. of Affected Individuals
Vitiligo	51	221
Autoimmune thyroid disease	46	119
Pernicious anemia	11	19
Adult-onset insulin-dependent diabetes mellitus	10	15
Addison disease	2	2
Systemic lupus erythematosus	1	1
Other autoimmune diseases	38	85

Note.—A total of 323 family members reported autoimmune diseases; some reported more than one autoimmune disease (mean 1.43 autoimmune diagnoses per individual). Not all individuals were genotyped.

Table 3

LODs (and *P* Values) on Phenotypic Stratification of 102 Multiplex Families with Vitiligo into Autoimmunity-Associated and Nonautoimmunity-Associated Family Subgroups

	LOD (*P*) FOR		
		Families with	
LOCUS	All Families (n = 102)	Autoimmunity (n = 51)	Nonautoimmunity (n = 51)
AIS1	5.59 (.000000279)	6.02 (.0000000887)	.22 (.158)
AIS2	3.73 (.0000208)	3.24 (.0000836)	.67 (.040)
AIS3	3.36 (.0000418)	.85 (.024)	2.88 (.000138)
SLEV1	3.07 (.0000852)	4.00 (.00000870)	.29 (.125)

pus and vitiligo (Nath et al. 2001), so the derivation of these linkage signals from the autoimmunity-associated family subgroup is not unexpected. In contrast, the *AIS3* linkage signal appears to derive principally from the nonautoimmunity-associated family subgroup. *AIS3* may thus predispose to vitiligo per se, rather than to an autoimmune diathesis, although, of course, the basis of generalized vitiligo even in these families might still be autoimmune in nature.

Heterogeneity testing did not quite exclude the possibility that *AIS1* might also contribute to vitiligo susceptibility in the nonautoimmune families, since the total LOD score did not decrease significantly (P = .084) when the autoimmune and nonautoimmune family subgroups are combined, compared with the LOD in the autoimmune families alone. This was also the case for *AIS2* and *AIS3* (P = .362 and .192, respectively). These results suggest the possibility of allelic heterogeneity at these loci between these two groups of families. However, for *SLEV1*, there was a significant (P = .018) decrease in the LOD score when the autoimmune and nonautoimmune family subgroups were combined, a result that suggests linkage to *SLEV1* in the autoimmune families and nonlinkage in the nonautoimmune families. Furthermore, although *SLEV1* was originally detected in multiplex families with lupus with at least one case of vitiligo (Nath et al. 2001), there was only a single case of possible lupus among all of our 51 autoimmune families. Thus, linkage to *SLEV1* in these families indicates that *SLEV1* confers susceptibility to a broader range of autoimmune diseases than just lupus and vitiligo.

Our data thus indicate that generalized vitiligo can be divided into at least two distinct phenotypic subcategories that apparently involve different loci or alleles. Vitiligo associated with a specific constellation of autoimmune diseases is linked with *AIS1, AIS2,* and *SLEV1,* whereas vitiligo unassociated with other autoimmune diseases is linked with *AIS3*. These findings begin to elucidate the genetic underpinnings of vitiligo and to dissect the contributions of individual loci to the vitiligo-associated autoimmune disease diathesis.

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Electronic-Database Information

URLs for data presented herein are as follows:

National Center for Biotechnology Information, http://www.ncbi.nlm.nih.gov/ (for the deCODE map and the STS map)
Online Mendelian Inheritance in Man (OMIM), http://www.ncbi.lnm.nih.gov/Omim (for vitiligo and APS2)

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