ELECTRONIC LETTER

Highly significant linkage to chromosome 3q13.31 for rhinitis and related allergic diseases

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Background: Allergic diseases such as asthma and rhinitis have closely related phenotypes and often occur with atopy. They show strong familial and intra-individual clustering, suggesting overlapping disease aetiology. Various loci and candidate genes have been suggested to underlie allergy. Many or all are still inconclusive. Following genome-wide scans on multiple phenotypes, we previously suggested that chromosome 3q13.12–q21.2 harbours an allergy locus.

Objective: To identify candidate loci in the Danish population, two additional independent sets of sib-pair families were fine-scale mapped in candidate regions showing maximum likelihood scores (MLS) ≥ 1.5 in the genome-wide scans.

Results: Twenty eight microsatellite markers in a denser map on chromosome 3q were analysed in 236 allergy sib-pair families including 125 sib pairs with rhinitis. We report significant evidence for linkage to chromosome 3q13.31 for rhinitis (MLS 5.55, identity by descent (IBD) 63.9%) and atopy (increased specific immunoglobulin E) (MLS 3.71, IBD 61.7%). We obtained an MLS of 5.1 (IBD 67.3%) at 3q13.31 when sib pairs with both rhinitis and atopy were analysed.

Conclusion: This study reports the first statistically significant evidence for a genetic susceptibility locus for rhinitis and to our knowledge shows the most significant evidence to date of linkage for any allergy phenotype.

The prevalence of allergic diseases has increased during the last generation^{1 2} and there is little doubt that genetic as well as environmental factors contribute to the manifestation of allergic diseases.³

Rhinitis (OMIM 607154) and asthma (OMIM 600807) have closely related phenotypes and are often seen with atopy. They show strong familial and intra-individual clustering, suggesting overlapping disease aetiology, which complicates studying only one phenotype at a time. Atopy (OMIM 147050) is defined as a genetic predisposition for the development of an immunoglobulin E (IgE) mediated response to allergens. It is a complex disease with major genetic influence, characterised by genetic heterogeneity and polygenic inheritance. Most likely interactions between multiple genes and various environmental exposures lead to allergic disease.

The genetic model for allergic diseases remains to be determined, but a combination of general allergy genes and more phenotype specific genes has been hypothesised.⁴ Although multiple linkage and association studies have suggested many allergy loci, only very few studies have found allergy loci that meet criteria for genome-wide significance.⁵ No significant evidence for linkage to rhinitis has been shown so far, although significant evidence for linkage

to various allergic diseases has been shown on chromosomes 3q21, 5q31–q33, 7p14–p15, 14q24, and 20p13.^{6–10}

A two step procedure examining three Danish samples has been employed: a genome-wide scan in a limited number of families (sample 1), followed by fine-scale mapping in a much larger number of families from two additional sets of sib-pair families in candidate regions showing a maximum likelihood score (MLS) ≥ 1.5 in the genome-wide scans. The results from the genome-wide scans have been published previously^{11–13} as have results for some of the fine-scale mapping for a minor part of the patients included in the present study.¹⁴ Chromosome 3q was indicated as a candidate region by genome-wide scans on multiple phenotypes in step one of our two step procedure (using sample 1). This study reports linkage results from step two on chromosome 3q13.12–q21.2 using rhinitis, atopy, and asthma as phenotypes.

METHODS

Participants

The three samples in the present study consisted of a total of 236 Danish allergy families obtained from two different allergy studies. Atopy is defined as a positive reaction to at least one of the tested allergen specific IgE.

Samples 1 and 3

Samples 1 and 3 were ascertained for clinical allergy in the ITA (Inheritance of Type I Allergy) study^{11 12} and included 51 independent sib pairs with rhinitis (fig 1) from 43 sib-pair families. Sib pairs in sample 1 were used in the original genome-wide scan and were characterised by having a positive specific IgE in addition to possible additional allergic phenotypes, while sample 3 included sib pairs with multiple allergic phenotypes but no increased specific IgE. The comorbidity between rhinitis and asthma in these samples was very high: 76% of the probands in the rhinitis sib pairs also had asthma, and 64% of the sib pairs with rhinitis were also sib pairs with asthma. Allergic rhinitis was doctor diagnosed using standard criteria.¹⁵ Clinical asthma was diagnosed according to standard criteria¹⁶ and those with atopic asthma were diagnosed as having clinical asthma and a positive specific IgE measurement to at least one of 11 tested allergens (CAP RAST FEIA, Pharmacia, Uppsala, Sweden). The allergens tested were grass, birch, mugwort, olive, *Parietaria*, cat, dog, horse, mite (*Dermatophagoides pteronyssinus* and *Dermatophagoides farinae*), and *Cladosporium herbarum*; measurements were considered elevated at ≥ 0.35 kUA/l ($\geq$ class 1). The phenotype definitions have been previously described.^{11 12}

Abbreviations: IBD, identity by descent; IgE, immunoglobulin E; MLS, maximum likelihood score

Figure 1 Number of independent sib pairs in the phenotypic groups of samples 1–3. Sample 1 includes 39 sib pairs with rhinitis (14+25), sample 3 includes 12 sib pairs with rhinitis (4+8), and sample 2 includes 74 sib pairs with rhinitis (30+44). The total number of independent sib pairs in samples 1 and 3 is 124. The circle with the thick line indicates sample 1 on whom the initial genome-wide scans were performed.

Sample 2

Sample 2 included 136 asthmatic sib-pair families. In addition to the asthma diagnosis, 74 families had sib pairs with rhinitis and 71 sib-pair families had sib pairs with positive specific IgE (fig 1). The asthma inclusion criteria for sample 2 were clinically based on recurrent cough, wheezing, and dyspnoea and a positive methacholine inhalation challenge identical to that used in a larger Danish study.¹⁷ A positive response was equivalent to a provocative concentration causing a 20% fall in forced expiratory volume in 1 s (FEV_1) <8 mg ml^{-1} . All family members filled in a questionnaire based on that used in the European Respiratory Health Survey.¹⁸ The rhinitis phenotype was self-reported in the questionnaire (affirmative answer to “Have you ever had allergy in the nose, including hay fever?”).

Subjects were categorised as having positive specific IgE tests if specific IgE measurements were $\geq$ class 1 for at least one of the specific allergens tested (the same as sample 1 except for olive and *Parietaria*) (CAP Phadiatop, Pharmacia & Upjohn, Copenhagen, Denmark).

Informed written consent was obtained from all families before participation. The study was approved by the ethics committee.

Genotyping

Genomic DNA was prepared from whole blood by standard methods. Twenty eight microsatellite markers were analysed in samples 1–3 to fine map a region on chromosome 3p14.2–q21.2 identified in a genome-wide scan (using sample 1 only) by Haagerup *et al.*^{11,12} These included D3S1300, D3S3635, D3S2406, D3S2465, D3S2459, D3S3045, D3S3675, D3S3526, D3S1310, D3S1278, D3S3670, D3S1579, D3S3529, D3S1575, D3S2457, D3S1558, D3S2460, D3S3649, D3S1303, D3S3569, D3S3607, D3S3657, D3S3696, D3S1549, D3S3612, D3S1569, D3S1744, and D3S3022. The average distance between the markers in the region showing evidence for linkage was 3.1 cM, and 1.3 cM in the region showing strongest evidence for linkage (D3S3045–D3S3607). Primer sequences were obtained from The GDB Human Genome Database (www.gdb.org). PCR was performed using amplification conditions similar to those described in The GDB Human Genome Database. PCR was performed in 6 μ l volumes in 96 well plates (including four control samples). The amplified products were separated on a 4% polyacrylamide gel using an ABI PRISM 377 DNA sequencer and analysed using the included software (Applied Biosystems, Naerum, Denmark). To facilitate combined analysis between the three samples, allele designations were consistent throughout all samples.

Statistical analyses

The data from the affected sib-pair families were analysed by non-parametric multipoint linkage analysis using the MLS approach. The computer program MAPMAKER/SIBS¹⁹ was used for calculation of multipoint maximum likelihood identity by descent (IBD) allowing for dominance variance. An increment step setting of five facilitated computing of MLS scores between each marker. If a family consisted of more than one affected sib pair, only independent pairs were used for calculations. Raw genotype data were pooled when combined samples were analysed. Values for locus specific sibling relative risk ratio (λ_s) for the susceptibility locus at peak MLS were calculated from the z_0 parameter ($\lambda_s = 0.25/z_0$).²⁰

To test for genetic heterogeneity between the three samples of sib-pair families in the study, we used the predivided sample test approach.^{21,22} We compared the maximum LOD score over the entire dataset ($Z_{\max}^{\text{TOT}}$) with the sum of the $Z_{\max}$ values in the individual subgroups ($\sum_{i=1}^n Z_{\max}^{(i)}$), where n indicates the number of subgroups (for example, $n=3$). Under certain regularity conditions, the statistic $L = 2\ln 10 (\sum Z_{\max}^{(i)} - Z_{\max}^{\text{TOT}})$ is asymptotically distributed as a χ^2 with degrees of freedom equal to one less than the number of subgroups.²³

RESULTS

We fine mapped chromosome 3q14.2–q21.2 using 28 microsatellite markers in three independent Danish samples consisting of a total of 236 families with clinical allergy.

Figure 2A shows the results for rhinitis from multipoint analyses on samples 1 (39 sib pairs), 2 (74 sib pairs), 3 (12 sib pairs), and the three samples combined using MAPMAKER/SIBS. Analyses of the full sample resulted in highly significant evidence in favour of linkage (MLS 5.55) between marker D3S2457 and D3S1558 and IBD of 64.2% with average information content of 98% in the region. Analyses of the 74 affected sib-pair families of sample 2 alone revealed significant evidence for linkage to rhinitis (MLS 3.6) at marker D3S1558 and IBD of 64.7%. Samples 1 (39 sib pairs) and 3 (12 sib pairs) showed MLS scores of 1.0 and 0.9 at the

Figure 2 Maximum likelihood IBD (MLS) curves for (A) rhinitis, (B) asthma, and (C) atopy (positive specific IgE) showing results for the individual samples and for the samples combined. The black curve represents the combined sample, the dotted curve sample 1, the broken curve sample 2, and the grey curve sample 3. The following markers are genotyped but not included on the x axis due to lack of space: *D3S1278 and D3S3679; †D3S3529, D3S1575, and D3S2457; ‡D3S3649. Furthermore, D3S2457, which maps between the markers in group †, and D3S1558 are left out. Sample 3 does not have sib pairs with positive specific IgE. The grey bar represents 10 cM.

same position, respectively. Both samples 1 and 3 had higher MLS proximal to the maximum MLS, for samples 1 and 3 at D3S3670 (MLS 1.5) and D3S3529 (MLS 1.3), respectively. Results from the linkage analyses of the region showing strongest evidence for linkage for the full sample are shown in table 1. The MLS-1 region spans 6.4 cM. Locus specific sibling relative risk ratio (λ_s) for the susceptibility locus at peak MLS was 1.4 for the combined sample.

The predivided sample test was used to test for heterogeneity between the three independent sib-pair samples with rhinitis and gave $L = 2\ln 10((3.6+1.0+0.9)-5.43) = 0.32$, which is not significant for a χ^2 with 2 degrees of freedom (which requires $\chi^2 = 5.99$ at the 5% significance level). The test was carried out for MLS values from the marker closest to maximum MLS, D3S1558.

An overlapping set of 194 pedigrees including 206 independent sib pairs with asthma was also analysed. Figure 2B shows evidence for increased IBD sharing for asthma sib pairs in the same region as shown for rhinitis. The linkage analyses did, however, result in a lower MLS (4.06, IBD 59.6% for all samples combined) at the marker D3S3670 despite the fact that more sibs were included in the analyses compared to rhinitis. Increased IBD sharing was also seen just proximal to the peak obtained using rhinitis as phenotype. This region showed suggestive evidence for linkage (MLS 2.76, IBD 59.4%) for the full sample between D3S2459 and D3S3045.

Since most of the offspring affected with rhinitis also had asthma (76% in samples 1 and 3 and 100% in sample 2), it was not possible to determine if the increased IBD sharing was due to a risk gene for rhinitis or if the region contained a gene causing a more severe phenotype including both asthma and rhinitis in the same patients. In an attempt to distinguish between the two phenotypes, we analysed 99 sib pairs with asthma but not rhinitis. This gave no indication of linkage at the peak seen from the rhinitis results (MLS 0.35, IBD 55.2%), but weak evidence for increased IBD sharing more proximal at D3S2406 (MLS 1.97, IBD 61.3%) (table 2).

Increased total IgE was the phenotype showing the strongest evidence for linkage in the original genome-wide scan (MLS 2.41),¹² however, this phenotype was not available for sample 2 and instead a positive specific IgE test was used as a measure for atopy in our study.

Figure 2C shows the result of the linkage analyses using atopy as phenotype and shows MLS values of 3.26 (IBD 60.5%) and 3.71 (IBD 61.7%) at D3S3670 and D3S1303, respectively, using a total of 130 sib pairs. MLS at the locus showing the best evidence for linkage for rhinitis was 2.55 for atopy. Locus specific sibling relative risk ratios (λ_s) for the susceptibility loci at peak MLS were 1.3 for both peaks in the combined sample.

In contrast, an increase in IBD sharing was observed when pedigrees with a positive specific IgE test and rhinitis were analysed (IBD 67.3%, MLS 5.07 between D3S3670 and D3S1579) for a total of 83 sib pairs (table 2).

DISCUSSION

In this study we investigated evidence for an allergy susceptibility locus at chromosome 3q14.2–q21.2 in a Danish population. Preliminary results showed weak evidence for linkage to this region from genome-wide scans performed on part of the samples,^{11 12} and in the present study we fine-scale mapped the region with a denser set of markers in the full set of families, including 186 additional independent sib-pair families. Results from fine mapping a minor part of the sample with fewer markers have been published previously.¹⁴

Several other candidate regions of interest were indicated by the genome-wide scans, but the region on chromosome 3q showed evidence for linkage to multiple allergy phenotypes.^{11 12}

Analysis using patients with rhinitis resulted in the most significant finding (MLS 5.55) at 3q13.31. This result can be regarded as highly significant evidence for linkage ($p < 3 \times 10^{-7}$), according to criteria proposed by Lander and Kruglyak.⁵ However, these criteria are based on data from genome scans and are thus too conservative for a design like

Table 1 MLS for rhinitis and atopy

Cytogenetic position	Marker map positions (cM)	Marker map positions (Mb)	Marker	Info* rhinitis	MLS rhinitis	IBD rhinitis	Info atopy	MLS atopy	IBD atopy
3p13	102.64	73.341	D3S2406	0.64	0.30	54.2	0.62	0.21	53.4
3p12.1	111.89	84.749	D3S2465	0.74	0.72	56.2	0.71	0.70	55.7
3q12.3	119.09	103.658	D3S2459	0.90	1.02	56.7	0.89	0.72	55.1
3q13.12	124.16	108.473	D3S3045	0.94	1.34	57.0	0.92	0.74	55.0
3q13.2	127.89	114.329	D3S3675	0.98	2.42	59.1	0.98	1.55	57.1
3q13.31	129.11	115.968	D3S3526	0.98	3.75	61.5	0.97	2.23	58.6
	129.73	116.265	D3S1310	0.99	4.09	61.9	0.99	2.71	59.5
	129.73	116.606	D3S1278	0.98	4.69	62.9	0.98	2.92	59.5
	129.73	116.644	D3S3670	0.98	5.11	63.5	0.98	3.26	60.5
	131.83	117.423	D3S1579	0.99	4.76	63.0	0.98	3.25	60.5
	131.83	117.556	D3S3529	0.99	5.20	63.6	0.98	3.28	60.2
	131.83	117.672	D3S1575	0.99	4.83	63.1	0.98	2.98	60.2
		118.674	D3S2457	0.97	5.46	64.0	0.96	2.86	60.0
				0.96	5.55†	64.2			
	133.93	118.733	D3S1558	0.99	5.43	63.9	0.99	2.55	59.7
3q13.31	134.64	118.885	D3S2460	0.99	5.22	63.6	0.98	2.97	60.2
3q13.32	134.64	119.127	D3S3649	0.99	5.22	63.6	0.98	2.96	60.2
	136.32	119.641	D3S1303	0.97	3.98	62.0	0.96	3.71†	61.7
3q21.2	140.19	126.300	D3S3569	0.87	2.01	58.7	0.86	1.68	59.2

Detailed results for the region showing evidence for linkage for the total sample. Atopy is defined as a positive specific IgE test. Genetic positions for markers were obtained from the Marshfield Center for Medical Genetics, and cytogenetic and physical positions from the UCSC Genome Browser, May 2004 assembly.

*Information content at the specific point; †highest maximum likelihood score (MLS) for each phenotype.

ours since most of our sib-pair families are only tested in a candidate region.

The fact that the three samples were not ascertained using exactly the same protocol could be regarded as a weakness causing the study to have decreased power, but this does not influence the significance of the findings. No genetic heterogeneity was found between the three samples using the predivided sample test.

Significant MLS scores were observed for rhinitis over a region of 7 cM, approximately the same size as the region defined by a decrease of 1 in MLS. Even though the validity of self reported rhinitis could be questioned, the results show that a sufficiently homogeneous aetiological group of patients was defined, as it would not be possible to obtain significant linkage otherwise.

Linkage data from sib pairs with asthma and/or atopy (positive specific IgE test) are also presented to facilitate discussion of aetiological overlap between the closely related phenotypes.

Using asthma as a phenotype gave evidence for linkage in the same position as observed using rhinitis and, furthermore, an additional region showing increased MLS was observed. However, the evidence for both asthma peaks is weaker than the evidence using rhinitis despite the fact that more sib pairs were included; if sib pairs with asthma but not rhinitis were used no or only marginal evidence for linkage was obtained. This could indicate that chromosome 3q13.31 harbours a gene that mediates an increased risk of developing rhinitis or possible severity of disease since most of the rhinitis sib pairs also have asthma. Due to the ascertainment scheme especially in sample 2 (all sib pairs sampled due to their asthmatic status), we are unable to determine if the region harbours a pure rhinitis locus since we have very few sib pairs with rhinitis but without asthma.

The initial genome-wide scan showed the most convincing linkage results for chromosome 3q13.32–q22.3 when using patients with increased total IgE levels.¹² Total IgE has not been measured in sample 2, but since all samples were tested

Table 2 MLS for mixed phenotypes

Cytogenetic position	Marker map positions (cM)	Marker	Info asthma – rhinitis	MLS asthma – rhinitis	IBD asthma – rhinitis	Info rhinitis + atopy	MLS rhinitis + atopy	IBD rhinitis + atopy
3p13	102.64	D3S2406	0.72	1.97*	61.3	0.60	–	–
3p12.1	111.89	D3S2465	0.75	1.76	60.6	0.70	–	–
3q12.3	119.09	D3S2459	0.88	1.59	60.4	0.88	–	–
3q13.12	124.16	D3S3045	0.91	–	58.4	0.92	1.63	59.6
3q13.2	127.89	D3S3675	0.97	–	56.8	0.96	2.85	62.5
3q13.31	129.11	D3S3526	0.98	–	54.1	0.97	3.59	64.1
	129.73	D3S1310	0.99	–	54.1	0.98	4.17	65.2
	129.73	D3S1278	0.99	–	54.0	0.97	4.48	65.9
	129.73	D3S3670	0.97	–	54.6	0.98	4.98	66.8
						0.94	5.07*	67.3
	131.83	D3S1579	0.98	–	56.1	0.99	4.55	65.9
	131.83	D3S3529	0.98	–	56.1	0.99	4.58	66.0
	131.83	D3S1575	0.97	–	55.5	0.99	4.14	65.1
		D3S2457	0.96	–	55.7	0.98	4.29	65.3
	133.93	D3S1558	0.98	–	54.2	0.99	4.27	65.3
3q13.31	134.64	D3S2460	0.97	–	53.8	0.99	4.64	66.1
3q13.32	134.64	D3S3649	0.97	–	53.7	0.99	4.67	66.1
						0.97	4.82	66.7
	136.32	D3S1303	0.94	–	52.9	0.97	4.77	66.5
3q21.2	140.19	D3S3569	0.86	–	50.2	0.86	1.78	61.1

Detailed results for mixed phenotypes using the total samples, which include 84 sib pairs with both rhinitis and atopy (positive specific IgE) and 94 pedigrees with asthma but not rhinitis. MLS scores <1.0 are indicated by –.

*Highest MLS for each phenotype.

for specific IgE, we used a positive specific IgE test as a measure for atopy. Linkage analyses using these patients suggested evidence for linkage (MLS 3.71), which supports the hypothesis of chromosome 3q13.31–q13.32 harbouring a susceptibility gene for atopy. In order to establish if the locus was involved in the atopy phenotype and determine if the evidence for linkage was not only due to the included rhinitis sib pairs, we excluded the families with rhinitis sib pairs from this group. This resulted in a decrease in IBD sharing to 55% ($n_{\text{fam}} = 41$, MLS 0.16) in the region that had previously demonstrated linkage, again suggesting that the locus contains a gene involved in the development of rhinitis.

In order to examine if objective measurements of atopy would support the self reported rhinitis diagnosis, we analysed the subset of rhinitis sib pairs who also had a positive specific IgE test and observed that the IBD sharing increased to 67.3% for the total sample. This indicates that inclusion of a positive specific IgE test in the phenotype might define an aetiologically more homogeneous group of patients.

Most genome-wide scans using allergy phenotypes have been performed on asthma and none of them have shown significant linkage to chromosome 3q, which is in keeping with our results, indicating that the high risk gene on 3q is involved with allergy phenotypes other than asthma. Few studies have examined linkage to rhinitis, but Yokouchi *et al* reported results from a genome-wide scan on seasonal allergic rhinitis with no linkage to 3q.²⁴ The same is reported by Dizier and co-workers in a recent paper on patients with rhinitis and asthma.²⁵

The region on 3q has, however, been suggested as a susceptibility locus for atopic dermatitis by several groups,^{6–26} for positive skin prick tests,²⁷ and for mite sensitisation.²⁸ Lee *et al* presented significant evidence for linkage to chromosome 3q21 mapping to marker D3S3606, 6–8 cM distal to our locus, using both atopic dermatitis and increased total IgE and stated that the data indicated that distinct genetic factors predispose to atopic dermatitis.⁶ A new paper by Kurz *et al* reports suggestive evidence for linkage to chromosome 3q21 using sib pairs sensitive to mite (*D pteronyssinus*) for the same marker as that showing maximum linkage in the study by Lee *et al*.²⁸ We only obtained an MLS of 1.43, 0.97, and 0.75 for marker D3S3607 (which maps to the same position as D3S3606, Marshfield Center for Medical Genetics) for rhinitis, asthma, and positive specific IgE, respectively. One explanation for these conflicting results could be that two adjacent loci map to chromosome 3q, the most proximal causing rhinitis and the distal causing atopic dermatitis/atopy. An alternative explanation is that chromosome 3q harbours one gene involved the general allergy process, even though the two regions on 3q do not overlap, as variation in location of linkage can vary substantially between replication studies, even when using relatively large numbers of families.²⁹ The involvement of common susceptibility genes in immune diseases has been suggested by several reports.^{30–31}

No obvious functional allergy candidate genes map to the region marked by a decrease of 1 in maximum MLS for rhinitis (<http://www.ncbi.nlm.nih.gov/mapview>, build 35.1) even though a number of known and unknown genes map to the region. Several candidate genes map to the broader candidate region between D3S2459 and D3S3569 and seem interesting with regard to the allergic process. These include ALCAM (activated leukocyte cell adhesion molecule, OMIM 601662), CD47 (CD47 antigen, OMIM 601028), TRIM (T-cell receptor interacting molecule, OMIM 604962), TACTILE (CD96, T cell activation, increased late expression, OMIM 606037), MOX2 (membrane glycoprotein MRC OX-2, OMIM 155970), and IGSF11 (immunoglobulin superfamily, member 11, OMIM 608351), not to mention all the genes

with still unknown function. Since 137 genes are listed in the region D3S2459–D3S3569 (<http://www.ncbi.nlm.nih.gov/mapview>, build 35.1), linkage disequilibrium mapping is needed to further define the candidate region. CD80 and CD86 have previously been suggested as candidates for chromosome 3q,⁶ however, they map approximately 2 and 4.5 Mb distal to the region that showed strongest evidence for linkage in our study.

In summary, we have obtained the first statistically significant evidence for the location of a susceptibility gene for rhinitis and this study shows, to our knowledge, the strongest evidence for linkage reported in the literature to date for any allergy phenotype. The gene could be involved in allergy severity but does not seem to be involved in the development of asthma.

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ELECTRONIC-DATABASE INFORMATION

The GDB Human Genome Database web site is at www.gdb.org and the NCBI Map Viewer can be found at <http://www.ncbi.nlm.nih.gov/mapview>. The web site of the Marshfield Center for Medical Genetics is at <http://research.marshfieldclinic.org/genetics/>.

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