Systemic lupus erythematosus (SLE) is an autoimmune disease with diverse and variable clinical manifestations and unknown etiology. Epidemiological and animal studies indicate that environmental and genetic factors are involved in the development of the disease. Several candidate gene loci (including the human leucocyte antigen (HLA) region, Fcγ receptors, and complement components) have been implicated through association studies, and multiple susceptibility loci have been detected in inbred mouse models of SLE.1,2 Until now, six groups have published genomewide scans with SLE as a phenotype in different ethnic groups.3–8 Recently, linkage to chromosome 2q37 (logarithm of odds (LOD) 4.24) in a Swedish population resulted in the identification of a new susceptibility gene PDCD1 in a large multinational study by Prokunina et al.9,10 The SLE associated allele of this immunoreceptor gene alters a binding site for the runt related transcription factor 1 (RUNX1), which is found in an intronic enhancer.

Stratification of pedigrees based on clinical manifestations has been used in recent studies that involved genomewide scans.11–16 The aim was to achieve genetically and clinically homogeneous sets of families and to increase the power to detect susceptibility genes for different subphenotypes of SLE. Altogether, 17 regions have been linked significantly to SLE with model based and non-parametric approaches; 11 of these in stratified studies. In addition, several other regions with suggestive linkage have been identified, but only some of those loci have been implicated in more than one study.13–18

The data suggest that multiple genes are involved in conferring susceptibility to SLE.

In our study, we conducted a nationwide and genomewide scan for SLE susceptibility loci in Finnish families multiply affected by SLE. The extensive hospital registration system in Finland allowed us to identify and recruit approximately 85% of all patients with SLE who needed hospital based treatment. Among these patients, we identified 35 multiplex families suitable for linkage mapping. We genotyped DNA samples from 73 patients with SLE and 96 healthy relatives with polymorphic microsatellite markers and analysed them with non-parametric linkage analysis. We obtained suggestive evidence for linkage (that is, non-parametric linkage scores that exceeded the threshold of one random occurrence per genome scan19) for three regions in a region on chromosome 14q with a previous suggestive mapping result,7 a region on chromosome 6q25–q27 previously linked to insulin dependent diabetes and rheumatoid arthritis, and a new locus on chromosome 5p. In addition, the HLA region on chromosome 6p had positive linkage that reached the suggestive threshold after a marker gap was filled. Our genome scan identified regions for linkage disequilibrium mapping in a larger cohort of Finnish patients with SLE and control participants.

**PARTICIPANTS AND METHODS**

**Patient recruitment**

The patients were recruited as described previously.19 The study was approved by the local ethical committees according to applicable regulations. Patients treated at Kuopio and Helsinki University hospitals, patients registered by the Lupus Foundation of Finland, and patients who answered an advertisement published in patient bulletins were recruited in 1995. We identified patients who had been treated in the two university hospitals during 1992–1995 from the corresponding hospital registries and contacted all patients personally or by mail. A second recruitment phase started in autumn 1996. We contacted doctors (mainly rheumatologists) from 17 central hospitals in Finland and four other major hospitals that treated patients with SLE. A letter was sent to all patients with the clinical diagnosis of SLE who had been treated in these hospitals during 1993–1996. All patients were asked whether they had relatives or family members diagnosed with SLE or a connective tissue disease similar to SLE. We asked patients with a positive family history to participate and obtained informed consent. According to the prevalence of SLE in Finland,20 we succeeded in contacting roughly 1200 out of 1500 available patients personally (by phone or by mail) during the two phases of recruitment; this accounted for about 85% of all Finnish patients with SLE who needed...
hypothesis of no linkage. The simulations consisted of 200 iterations, as described previously. In each iteration, we drew founder alleles from the estimated allele frequency distributions and introduced missing genotypes to exactly the same positions as in the real data. As a result, we obtained an empirical distribution for the non-parametric linkage scores, which corresponded to suggestive and significant linkage observed under the null hypothesis in the entire genome scan.

RESULTS

Demographic, clinical, and laboratory features of families with SLE

Seventy-three patients with SLE and 96 healthy relatives from 35 multiply affected families were identified. Their clinical features have been described previously. In 32/35 families, two family members were affected. The most common combination was sister and sister (16 families); this was followed by aunt and niece (eight families). Three families had three patients with SLE. In two of these families, the affected family members were first degree relatives; in the third family, a second degree relative (a cousin) and first degree relatives (mother and son) were involved. Seven (9.6%) of the 73 familial cases were men, which corresponded well with the sex distribution of SLE in the general population.

Genome scan

We used the marker set MD-10 to capture 50–60% of the maximum inheritance information throughout the genome. Loss of information was mainly caused by the pedigree structure (that is, missing parents). Table 1 shows the highest non-parametric linkage scores obtained and the corresponding information content (chromosome 3, chromosome 5, and chromosome 6).

We used simulations to establish the empirical thresholds for suggestive and significant linkage (that is, non-parametric linkage scores reached at random once per one or

<table>
<thead>
<tr>
<th>Chromosome</th>
<th>Marker</th>
<th>Non-parametric linkage score</th>
<th>p value</th>
<th>Information content</th>
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<tbody>
<tr>
<td>3q</td>
<td>D3S1278</td>
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<td>0.64</td>
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<td>8</td>
<td>D8S260</td>
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<td>14q</td>
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<td>0.02</td>
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</table>
20 genome scans, respectively). The best observed non-parametric linkage score under the null hypothesis of no linkage showed that non-parametric linkage = 1.7 is reached once per genome scan (suggestive linkage) and that the global p value = 0.05 corresponds to non-parametric linkage = 3 (significant linkage). Three loci exceeded the threshold for suggestive linkage; none was significant (table 1). To verify the highest non-parametric linkage scores and to cover gaps in the map, we genotyped more markers at regions with non-parametric scores >1.7 (chromosome 3q, chromosome 5p, and chromosome 6q) and regions with gaps >20 cM if the corresponding non-parametric linkage score was >1 (chromosome 6p (that is the HLA region), chromosome 8, and chromosome 14). In all regions with suggestive

![Figure 1](image-url)
linkage, non-parametric linkage scores increased with additional markers (table 2). The most striking change in non-parametric linkage scores was seen in the HLA region, in which the non-parametric linkage score increased from 1.2 to 2.1. Figure 1 shows the overall non-parametric linkage scores, including all genotyped markers.

DISCUSSION

This was the first nationwide study in Finland to map genetic factors that confer susceptibility to SLE. We recruited >80% of patients with SLE in Finland, which has a population of 5 million. Our results suggest that no single major gene contributes to the risk of the disease in Finland, despite its population structure. Our genome scan showed three loci that exceeded the threshold of 1.7; this corresponded to the definition of suggestive linkage genomewide. Even after we increased the amount of information with additional markers, no locus reached the threshold for significant linkage. Ideally, the results should be verified in an independent dataset. Unfortunately, we cannot increase the linkage.

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Chromosome 14q21–q23 also has been implicated repeatedly in previous linkage studies, although no study alone has reported a significant linkage result. Evidence for linkage to chromosome 14q has been reported in three independent data sets: Gaffney and colleagues (LOD 2.81, p = 0.0016), Shai and colleagues (non-parametric linkage 2.02, p = 0.02) and Lindqvist and colleagues in patients of Swedish pedigree. The HLA region also has been implicated in more recent linkage mapping studies. Gaffney and colleagues reported strong linkage to the HLA region; this was supported by results from Shai and colleagues and Lindqvist and colleagues in patients of Swedish pedigree.5 7

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The biological relevance of the observed excess sharing of alleles in the novel regions on chromosomes 5p and 6q4 is more difficult to evaluate. Interestingly, the same region on chromosome 6q25–q27 has been implicated in other autoimmune diseases. Several studies support a locus that confers susceptibility to insulin dependent diabetes in this chromosomal region (IDDM5).21 22 Supportive evidence for the existence of a common autoimmune susceptibility locus in the region comes from a study by Myerscough and colleagues.23 They report evidence for linkage disequilibrium between rheumatoid arthritis and the markers D6S311 and D6S440. As the highest observed Z score in our study resided in the same region, chromosome 6q25–q27 may harbour a gene that influences the predisposition to autoimmunity.

Recently, Rioux and colleagues showed the power of linkage disequilibrium analysis in complex disease mapping.24 By genotyping polymorphic markers at 0.35 cM intervals in a region initially identified by linkage mapping, they successfully identified linkage disequilibrium and an ancestral haplotype that spanned 250 kb in patients with inflammatory bowel disease in an outbred Canadian population. A similar approach should be feasible among Finnish patients and controls in an expanded study. In a population characterised by local founder effects, linkage disequilibrium mapping in selected geographical areas might be a useful strategy for identifying relatively rare susceptibility alleles.25 We intend to use the power of linkage disequilibrium to identify common ancestral chromosomes among a larger cohort of patients with SLE and controls by high resolution mapping.

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REFERENCES

Linkage mapping of systemic lupus erythematosus in Finnish multiplex families


O’Connell JR, Weeks DE. PediCheck: a program for identification of genotype.


