Am. J. Hum. Genet. 55:1230–1241, 1994

Analysis of HLA and Disease Susceptibility: Chromosome 6 Genes and Sex Influence Long-QT Phenotype

Lowell R. Weitkamp,[1] Arthur J. Moss,[2] Raymond A. Lewis,[5] W. J. Hall,[3] Jean W. MacCluer,[6] Peter J. Schwartz,[7] Emanuela H. Locati,[8] Dan Tzivoni,[9] G. Michael Vincent,[10] Jennifer L. Robinson,[4] and Sally A. Guttormsen[1]

[1]Division of Genetics, [2]Department of Medicine, [3]Department of Biostatistics, and [4]Department of Community and Preventive Medicine, University of Rochester School of Medicine and Dentistry, Rochester; [5]Department of Physics, Pennsylvania State University, University Park; [6]Department of Genetics, Southwest Foundation for Biomedical Research, San Antonio; [7]Department of Medicine, University of Pavia, Pavia; [8]Instituto Clinica Medica 2, University of Milan, Milan; [9]Department of Cardiology, Shaare Zedek Medical Center, Jerusalem; and [10]Department of Medicine, University of Utah School of Medicine, Salt Lake City

Summary

The long-QT (LQT) syndrome is a genetically complex disorder that is characterized by syncope and fatal ventricular arrhythmias. LQT syndrome, as defined by a prolonged electrocardiographic QT interval, has a higher incidence in females than in males and does not exhibit Mendelian transmission patterns in all families. Among those families that are nearly consistent with Mendelian transmission, linkage between a locus for LQT syndrome and the H-*ras*-1 locus on the short arm of chromosome 11 has been reported in some families but not in others. Earlier analyses suggesting that LQT syndrome might be caused by a gene in the *HLA* region of chromosome 6 were not confirmed by standard linkage analyses. Here, we present an analysis of HLA haplotype sharing among affected pedigree members, showing an excess of haplotype sharing in a previously published Japanese pedigree and possibly also in 15 families of European descent. The haplotypes shared by affected individuals derive from both affected and unaffected parents. In an analysis of independent (unrelated) HLA haplotypes, we also found a nonrandom distribution of *HLA-DR* genes in LQT syndrome patients compared with controls, suggesting an association between the LQT phenotype and specific *HLA-DR* genes. Our data indicate that DR2 has a protective effect and, particularly in males, that DR7 may increase susceptibility to the LQT syndrome. Thus, LQT syndrome may be influenced by genes on chromosomes 11 and 6, possibly with a sex-specific effect. These results provide a model for an effect of *HLA*-region genes inherited from either parent on the expression of an illness that may be determined principally by alleles at loci not linked to *HLA*.

Received April 14, 1993; accepted for publication August 22, 1994.

Address for correspondence and reprints: Dr. Lowell R. Weitkamp, Division of Genetics, University of Rochester Medical Center, 601 Elmwood Avenue, Rochester, NY 14642.

0002-9297/94/5506-0021$02.00

Introduction

The long-QT (LQT) syndrome (McKusick 1992, p. 1137) is a familial, idiopathic, electrocardiographic disorder with delayed ventricular repolarization (QT prolongation). It was first identified by Romano (Romano et al. 1963; Romano 1965) and Ward (1964) as having a dominant pattern of inheritance (Romano-Ward syndrome). Affected individuals are at risk for syncope and fatal ventricular arrhythmias (Schwartz et al. 1975; Moss 1986). In a prospective longitudinal study of 328 families, probands (who usually received medical attention because of a syncopal episode during childhood) had a risk of postenrollment syncope or LQT-related death of 5.0% and 0.9% per year, respectively (Moss et al. 1991). These event rates among probands were considerably higher than those among affected relatives as conventionally defined by a QT interval corrected for heart rate (QTc; Bazett 1920) >0.44 s. Additional evidence for complexity in the genetic basis of the LQT syndrome is the higher prevalence in females than in males of a QTc interval >0.44 (Hashiba 1978, 1992; Moss et al. 1991; Vincent et al. 1992) and the difficulty in defining a clinical phenotype that fits Mendelian transmission patterns in all families (Hashiba 1978, 1992; Weitkamp et al. 1989). The considerable variability of the QTc interval at different times in the same individual contributes to this difficulty.

In a study of one large Utah kindred (Keating et al. 1991*a*) and six unrelated follow-up kindreds (Keating et al. 1991*b*), a locus for the LQT syndrome was linked to the H-*ras*-1 locus on the short arm of chromosome 11 (lod score 21.69), with no recombinants among 66 affected individuals in the seven kindreds. The authors hypothesized that LQT is genetically homogeneous. Subsequently, a 131-member Israeli kindred showed evidence for Mendelian transmission of a gene for LQT syndrome and no linkage of the *LQT* locus to the H-*ras*-1 locus (Kerem et al. 1992; Benhorin et al. 1993). Towbin et al. (1992) concluded that the LQT syndrome is genetically heterogeneous on the basis of their observation that 7/11 families that they studied showed linkage to the H-*ras*-1 locus and 4/11 did not. The same group that first reported linkage

Table 1

LQT Families in the HLA Study

Family	No. of Subjects	Location; Ethnic Background
CA001	17	New York State; German
CA002	9	Wales; Welsh
CA003	7	New York State; English
CA004	16	New York State; Italian, British, German
CA005-CA008	11 + 5	Utah; Danish, English
CA006	9	Italy; Italian
CA007-CA009	8 + 9	Israel; Sephardic Jewish
CA010	6	Italy; Italian
CA011	5	Italy; Italian
CA012	3	Israel; Ashkenazi Jewish
CA013	10	Spain; Spanish
CA014	6	New York State; Italian, Irish
CA015	13	Italy; Italian

of *LQT* to H-*ras*-1 has also invoked locus heterogeneity to explain the absence of linkage between chromosome 11p15.5 marker loci and a postulated *LQT* locus in two families (Curran et al. 1993).

Earlier investigations suggested that an HLA-linked chromosome 6 gene might cause the syndrome. Itoh et al. (1982) reported a Japanese family in which 10 members affected with Romano-Ward syndrome (LQT) had the same HLA haplotype. A subsequent analysis of this family and part of one new family (CA001; table 1), assuming a dominant mode of transmission, yielded a peak lod score of 3.68 for linkage between *LQT* and *HLA*, with one apparent recombinant (Weitkamp and Moss 1985). Melki et al. (1987) reported two families with a peak lod score of 0.66 at $\theta = 0$, which, when added to the prior data, gave a peak lod of 4.23 at $\theta = .04$. However, Giuffre et al. (1990) performed a multipoint lod score analysis of six families, using probes within and flanking the *HLA* region and excluded (lod score <-2.0) a locus for LQT syndrome from the region 1 cM centromeric of DRβ to 5 cM telomeric of pCH_6 (D6S10). Keating et al. (unpublished results cited in Keating et al. 1991*a*) also excluded linkage of *LQT* with the *HLA* loci in the large family that showed linkage of *LQT* with H-*ras*-1.

Here, using the same clinical criteria for affected as were used by Keating et al. (1991*a*, 1991*b*), we report an analysis of *HLA* gene sharing among affected individuals in the pedigree published by Itoh et al. (1982) and in 15 two-to-four-generation follow-up families (Weitkamp et al. 1989). An affected-pedigree-member analysis (PEDSCORE; Weitkamp and Lewis 1992), requiring no genetic model of disease, was used to evaluate the possibility that affected relatives had increased sharing of HLA haplotypes identical by descent (IBD). The follow-up families include sections from pedigrees that were reported to show tight linkage between a postulated *LQT* locus and the H-*ras*-1 locus and from pedigrees that showed lack of linkage between these loci. We also report an association analysis of HLA-DR allele frequencies in unrelated haplotypes (counting only once haplotypes IBD) in persons with LQT syndrome. We conclude that genes in the *HLA* region of chromosome 6 constitute one of the elements in the multifactorial etiology of a prolonged QT interval. These results provide evidence that the expression of phenotypes that are dominant or nearly dominant (as in the LQT syndrome) may be influenced by *HLA*-region genes *inherited from either parent.*

Subjects and Methods

Study Population and Clinical Evaluation

Fifteen two-to-four-generation families from 13 apparently unrelated kindreds in a large, prospective international study of LQT syndrome were selected for HLA typing. Details of the study population and clinical evaluation have been given by Moss et al. (1991). For the purposes of that study, an affected family member was defined as having a QTc, according to Bazett's (1920) formula, of >0.44 s.

Families were selected for the HLA study on the basis of willingness to participate and availability of the proband and at least one other affected family member. They were geographically and ethnically diverse (table 1). Two of the families, CA005 and CA008, are very distantly related members of the large Utah kindred, in which a locus for LQT syndrome was linked to the H-*ras*-1 gene (Keating et al. 1991*a*). Two other families, CA007 and CA009, are from the 131-member kindred that showed no linkage of an *LQT* locus to the H-*ras*-1 locus on chromosome 11 (Kerem et al. 1992; Benhorin et al. 1993). These families, though related as first cousins, could not be joined by HLA-typed ancestors and were treated as separate families for analysis. Another family, CA004, is the same family identified as K1756 by Curran et al. (1993) in the report of

no linkage of *LQT* to the H-*ras*-1 locus. The parents in CA006 are second cousins.

For purposes of analysis of the relationship of *HLA* genes to the LQT phenotype, we have used the clinical criteria of Keating et al. (1991*a*, 1991*b*). Specifically, diagnosis of LQT syndrome was based on (*a*) QTc $\geq$ 0.45 in persons with a history of recurrent syncope or documented polymorphous ventricular tachycardia and (*b*) QTc $\geq$ 0.47 in persons without symptoms. The analysis was divided into two phases, which were based on the inclusion of different electrocardiographic data in determining clinical diagnosis. For the initial analysis, QTc intervals were provided by one of us (J.L.R.) without knowledge of the genetic-typing results, and, in the case of individuals with multiple electrocardiograms, QTc intervals were taken from the earliest available tracing. In the final analysis, QTc intervals previously entered into the database from all prior electrocardiograms were considered. The number of additional QTc intervals varied from 0 (for 46 persons) to 17. The determination of QTc intervals for the final analysis was made by one of us (A.J.M.) without knowledge of HLA types of family members. For persons with multiple electrocardiograms, a diagnosis of affected was made (for the final analysis) if at least one electrocardiogram showed QTc $\geq$ 0.47 *and* if the mean QTc of all electrocardiograms was $\geq$0.46.

The 88 family members who had 1–17 repeat QTc determinations included 59 of the 63 persons scored as affected in the initial analysis and 29 of the remaining 71 persons. Among the latter 29 persons, 6 new persons scored as affected in the final analysis were CA001, IV-2 (female, initial QTc = 0.46) with one repeat QTc = 0.49; CA003, II-1 (male, initial QTc = 0.46) with a mean QTc = 0.465 and 7 of 16 repeat QTc $\geq$ 0.47; CA003, III-1 (female, initial QTc = 0.45) with repeat QTc = 0.45 and 0.47; CA005, I-3 (male, initial QTc = 0.43) with repeat QTc = 0.52 and 0.60; CA007, I-1 (male, initial QTc = 0.45) with repeat QTc = 0.47 and 0.48; and CA014, II-3 (female, initial QTc = 0.46) with repeat QTc = 0.42, 0.46, and 0.48. Two persons scored as affected in the initial analysis were eliminated from affected status by the same criteria. They were CA006, II-7 (male, initial QTc = 0.49) with a repeat QTc = 0.38, and CA013, II-7 (female, initial QTc = 0.48) with repeat QTc = 0.36 and 0.43.

Genetic Typing

During a 4-year period (May 1984–March 1988), blood samples were collected by antecubital venipuncture into Vacutainers containing acid citrate dextrose (formulation A; NIH) from 140 members of 13 kindreds. Of these, 134 samples (table 1) were typed for HLA antigens (A, B, C, and most for DR) in Rochester within 48 h, using fluorescein diacetate and ethidium bromide in the standard NIH microlymphocytotoxicity test (Bruning 1976; Darke 1976). Antigen assignment was based on the highest level of variants, or "splits," afforded by the reagents used for typing; the assignment of phenotypes was consistent with the standards developed at the Eighth International Histocompatibility Testing Workshop. Properdin factor B (BF) and glyoxalase (GLO) were typed as described elsewhere (Alper et al. 1972; Lamm et al. 1978). HLA haplotypes were constructed for each family member, on the basis of that person's parents or offspring.

Analysis

For the pedigree analysis, the probability distribution for each possible number of matches of IBD HLA haplotypes among family members affected with LQT syndrome was calculated using PEDSCORE (Weitkamp and Lewis 1992). Only family members scored as affected were included in the analysis. The probability, P, of the observed number of matches or any more extreme number of matches was taken directly from the probability distribution. To evaluate the statistical significance of the results for the 15 follow-up families taken together, we used the "median value chi square" statistic of Lancaster (1949). The statistic, denoted χ'^2_m, is defined as

$$\chi'^2_m = -2 \log_e \tfrac{1}{2}(P + P'), \text{ if } P' \neq 0$$
$$= 2 - 2 \log_e P, \text{ if } P' = 0,$$

where P is defined as above and P' is the probability of all numbers of matches greater than the observed number of matches. This statistic, which has approximately a χ^2 distribution with 2 df, permits the combining of probabilities arising from discrete distributions (Lancaster 1949). A standard contingency χ^2 test was used for the various association analyses.

Results

Analysis of the Pedigree Reported by Itoh et al. (1982)

The pedigree reported by Itoh et al. (1982) is the only LQT syndrome pedigree with published HLA data of which we are aware. A slightly revised version of this pedigree, which takes into account that the oldest of five offspring of the top-generation female had a (deceased) father different from that of the other four offspring (S. Itoh, personal communication), is shown in figure 1. Nine of the 10 affected persons had a QTc 0.49–0.65; no data were presented for the 10th person. All 10 affected family members had a history of syncope; 8 of the 10 were females.

When dominant transmission and complete penetrance of an allele causing the LQT syndrome are assumed, the peak lod score for linkage between *LQT* and *HLA* is 2.71 at $\theta = 0$ (Weitkamp and Moss 1985). The MZ twins were counted as a single genetic event, and the two HLA-typed unaffected offspring were included in the 10 offspring scored. (The lod score is the same for both the pedigree configuration reported by Itoh et al. [1982] and the revised

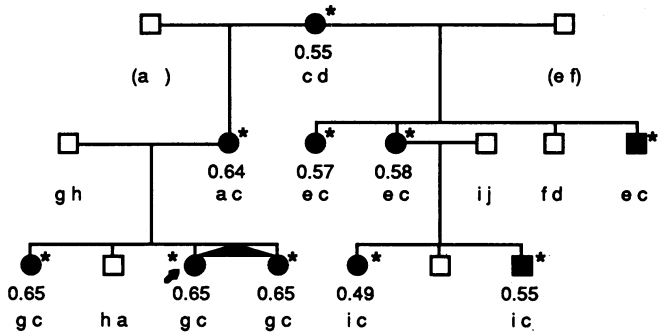

Figure 1 Revised version of the pedigree of the LQT family reported by Itoh et al. (1982) per Itoh (personal communication). The blackened symbols (circles denote females; and squares denote males) indicate a diagnosis of LQT syndrome, as reported by Itoh et al. (1982). QTc values and HLA haplotypes (*letters*) are shown below the symbols. An asterisk indicates a history of syncope. Inferred HLA haplotypes of untested persons are shown in parentheses. The pedigree was produced by Pedigree/Draw (Mamelka et al. 1990).

version shown here.) To calculate the probability that all of the HLA haplotype sharing observed among affected family members might have occurred by chance, we tabulated the relative probability for each possible number of pairwise IBD matches of HLA haplotypes from any ancestor. The results are shown in table 2.

For each of the eight affected offspring (counting the twins as one genetic event) there are four possible combinations of parental haplotypes. Thus, there are 4^8 (or 65,536) possible configurations of HLA haplotypes among the nine affected family members. Some pairs of relatives, e.g., parent and offspring, must, of course, share, or "match for," an HLA haplotype IBD. Other pairs of relatives, such as siblings and second- or third-degree relatives, may or may not match for an HLA haplotype IBD. The minimum possible number of pairwise IBD matches is 13; the maximum possible number of IBD matches is 41. The *relative* probabilities (of 4,096) of each of the possible number of matches shown in table 2 were generated by PEDSCORE (Weitkamp and Lewis 1992). The observed number of pairwise matches, obtained by manual counting, was 41. Thus, the probability, P, of the observed and any more extreme number of matches in this Japanese family was 2/4,096 ($p = .00049$).

Evaluation of LQT Syndrome in Family Members

HLA haplotypes and initial QTc intervals for the members of 15 follow-up families are shown in figure 2. The data differ from the prior tally (Weitkamp et al. 1989) in that blood samples received from six members of the CA006 kindred were erroneously counted as HLA typed, when they were not. These individuals are not shown on the pedigree in figure 2. Therefore, among individuals with an initial QTc $\geq$ 0.45, there were 83 HLA-typed persons, not 86, as previously reported. Twenty HLA-typed family members had a history of syncope, cardiac arrest, or polymorphous ventricular tachycardia; they are identified by an asterisk. All family members who were HLA typed were included in the pedigrees. Other individuals are shown only if they were necessary to clarify the genetic relationships of HLA-typed family members. Non–HLA-typed relatives who had a QTc $\geq$ 0.47 or a sudden, unexplained early death are listed in table 3. Kindred CA004 is the same family reported as K1756 by Curran et al. (1993); siblings are shown in the customary left-to-right birth order in figure 2.

Inspection of the pedigrees suggests an approximately dominant pattern of transmission of the LQT syndrome phenotype, using the criteria of Keating et al. (1991*a*, 1991*b*), namely QTc $\geq$ 0.47 in the absence of symptoms or QTc $\geq$ 0.45 with symptoms. This is apparent, on the basis of clinical status determined from the initial QTc (shown in fig. 2) and even more so when the eight revisions

Table 2

Relative Probabilities of the Number of IBD Pairwise HLA Haplotype Matches among Affected Persons in Pedigree in Figure 1

No. of IBD Haplotype Matches	Relative Probability (No. of Different Haplotype Configurations)
13	.00146 (6)
14	.01270 (52)
15	.03564 (146)
16	.07227 (296)
17	.10498 (430)
18	.10400 (426)
19	.09033 (370)
20	.10498 (430)
21	.11133 (456)
22	.07617 (312)
23	.05078 (208)
24	.04492 (184)
25	.04541 (186)
26	.04053 (166)
27	.02197 (90)
28	.01221 (50)
29	.00977 (40)
30	.01074 (44)
31	.01611 (66)
32	.01367 (56)
33	.00635 (26)
34	.00293 (12)
35	.00195 (8)
36	.00098 (4)
37	.00146 (6)
38	.00293 (12)
39	.00195 (8)
40	.00098 (4)
41	.00049 (2)
Total	.99999 (4,096)

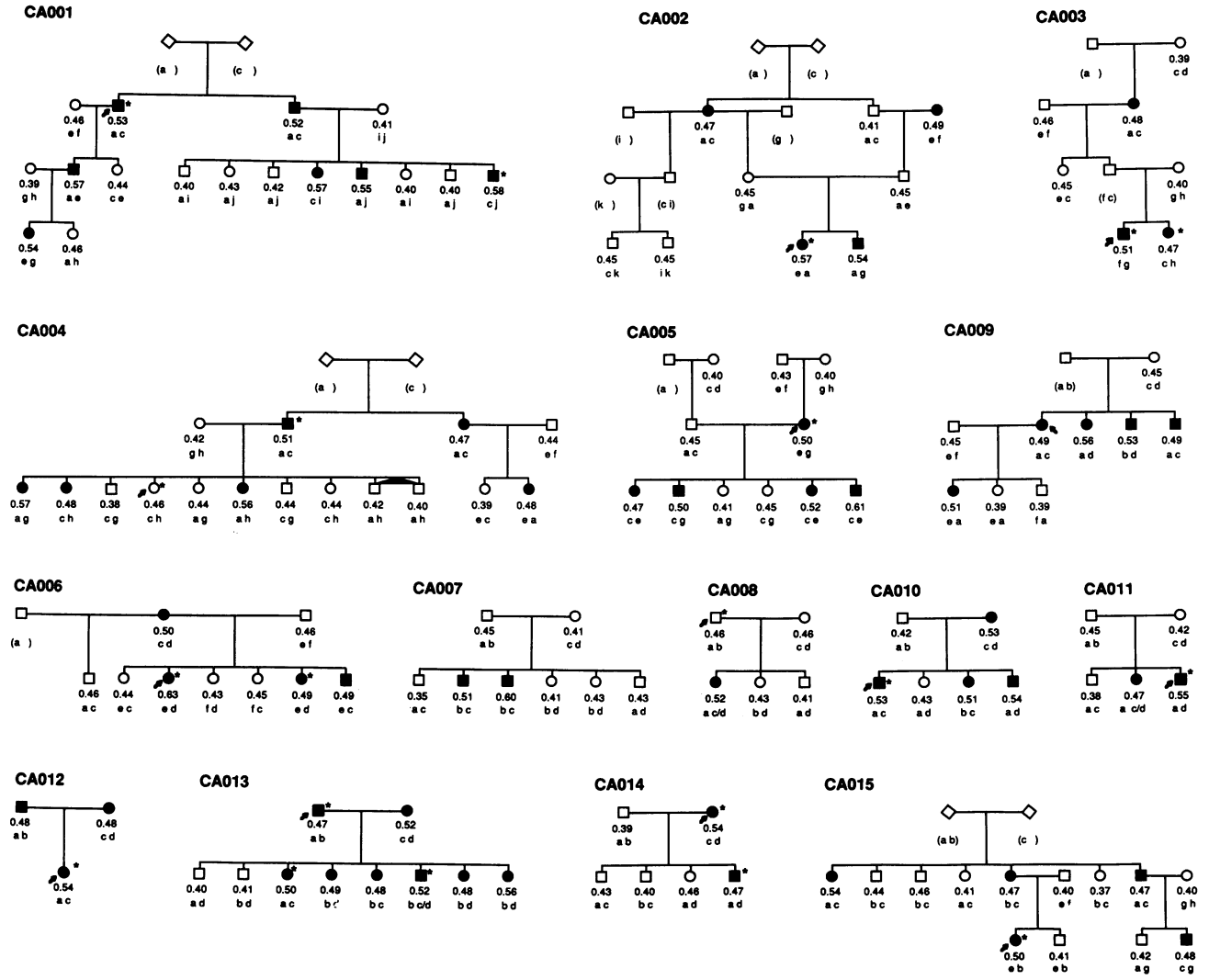

Figure 2 Fifteen follow-up LQT families, showing QTc interval and HLA haplotypes (letters). The blackened symbols (circles denote females; and squares denote males) indicate a QTc of ⩾.47 on the initial electrocardiogram. Probands are identified by an arrow; there is no arrow shown for CA007, as the male with QTc = .45 was ascertained by virtue of being a first cousin of the proband in CA009. CA005 and CA008 are very distantly related families from the same kindred. Only siblings from whom blood samples were obtained are shown on the pedigree. A history of syncope or cardiac arrest is indicated by an asterisk. In addition, the father of the proband in CA003 had a history of seizures or syncope and died suddenly at age 19 years. The QTc interval from the initial electrocardiogram and HLA haplotypes (*letters*) are shown below the pedigree symbol. The HLA recombinants are in CA008 between *HLA-B* (c) and *HLA-DR* (d), in CA011 between *HLA-A* (c) and *HLA-B* (d), and in CA013 between *HLA-B* (c) and *HLA-DR* (d). Inferred HLA haplotypes of untested persons are shown in parentheses. The pedigrees are correct representations and may be used by the reader in other analyses. Siblings are presented in birth order, left to right. The pedigrees were produced by Pedigree/Draw (Mamelka et al. 1990).

in status that were based on subsequent QTc values available for some family members are taken into account. Among 19 nuclear families in which both parents were evaluated and there was an affected offspring, there are 2 pedigrees (CA005 and CA007) in which neither parent was affected in the initial analysis but in which one parent was affected in the final analysis. In two other pedigrees in which neither parent was affected (CA002 and CA011), repeat QTc determinations were not available for the parents. Clearly, carriers of a major gene contributing to a long QT interval might have an occasional QTc < 0.46. In three families (CA003, CA012, and CA013) both parents were affected, on the basis of final diagnoses, and in five families (CA001, CA005, CA006, CA008, and CA009), the unaffected spouse of an affected parent had a QTc = 0.45 or 0.46. Such a large proportion of families having two parents with a long (⩾0.45) QTc interval (8/19) would be unlikely if this disorder were a simple autosomal dominant with occasional incomplete penetrance. It suggests that genes from both parents may contribute to the expression of the full LQT syndrome phenotype (criteria of Keating et al. 1991*a*, 1991*b*).

Table 3

Non-HLA Typed Relatives with QTc ≥0.47 or Sudden, Unexplained Early Death

Family	Individual (Sex, QTc)	Comments
CA001	III-5 (F, .43)	Daughter with QTc = .48
CA002	III-3 (F, .45)	Brother with QTc = .38; brother's daughter with QTc = .47
	III-4 (M, .45)	Brother with QTc = .43; brother's son with QTc = .47
CA003	III-2 (M, . . .)	He and sister had history of "seizures" and died suddenly at ages 19 and 16 years, respectively
CA004	II-2 (M, .51)	Brother (QTc = .51) died of "heart attack" at age 51 years
	III-4 (F, .46)	Sister died suddenly at age 15 years[a]
CA005		Many affected[b]
CA006	II-3 (F, .63)	Sister died suddenly at age 19 years
CA007		Many affected[c]
CA008		Many affected[b]
CA009		Many affected[c]
CA010	II-1 (M, .53)	Sister died suddenly at age 14 years
CA013	I-1 (M, .47)	Daughter, sister, niece, and nephew (QTc = .54) died suddenly at ages 1, 26, 9, and 15 years, respectively
	II-3 (F, .50)	Son with QTc = .53
CA014	I-2 (F, .54)	Mother and brother each with QTc = .51
CA015	II-2 (M, .44)	Daughter died suddenly at age 22 years

[a] Shown on pedigree reported by Curran et al. (1993).
[b] Shown on pedigree reported by Keating et al. (1991*a*).
[c] Shown on pedigree reported by Benhorin et al. (1993).

HLA Haplotype Sharing among Affected Family Members

The analysis of the family reported by Itoh et al. (1982) suggests that *HLA*-region genes IBD are shared among affected family members more often than is expected on a random basis. We tested this observation in the 15 follow-up families. The relative probability of each possible number of HLA haplotype matches IBD among affected members of each family was tabulated using PEDSCORE (data not shown), as in the example in table 2; DR genes were used to mark the HLA haplotype transmitted in the three intra-HLA recombinant individuals (fig. 2).

The probability P for the number of haplotype matches among affected family members that is greater than or equal to the observed number of haplotype matches is shown for each family, for both the initial and final analyses, in table 4. None of the families are large enough for an evaluation of the possibility of heterogeneity among families in the results. Thus, we combined the probabilities from each family, for a test of the statistical significance of the total follow-up data. In order to do this, we calculated the χ'^2_m (Lancaster 1949), which is distributed as a χ^2 with 2 df. The sum of χ'^2_m for the 12 informative families in the initial analysis was 32.56 (24 df; $p = .114$). A slightly more significant value ($p = .058$) was obtained in the analysis based on the final set of diagnoses (table 4).

In order to estimate whether increased HLA haplotype matching might have occurred in a subset of families that showed or did not show linkage between a locus for LQT syndrome and the H-*ras*-1 gene, we considered the two pedigrees that showed linkage (CA005 and CA008) separately from the three pedigrees (CA004, CA007, and CA009) that did not show linkage. The CA008 pedigree was uninformative, and, therefore, χ'^2_m for the H-*ras*-1–linked variety (final diagnoses; table 4) was 5.3096 ($p = .07$). For the three pedigrees not showing linkage between H-*ras*-1 and an LQT syndrome locus, the sum of χ'^2_m was 9.3687 (6 df, $p = .15$). Thus, we could not determine whether the HLA haplotype matching between affected members was limited to either subset of the families.

Association with HLA-DR Genes

To investigate the possibility that specific *HLA-DR* genes may be associated with the expression or lack of expression of the full LQT phenotype, we tabulated the *HLA-DR* genes that were present on haplotypes that (*a*) occurred in affected family members (final diagnoses), (*b*) did not occur in affected family members but did occur in persons with QTc = 0.45 or 0.46, and (*c*) occurred only in persons with QTc ≤ 0.44. There were 81 unrelated haplotypes in the 15 families, of which all but haplotype "a" in family CA006 were typed for *HLA-DR*. The results are presented in table 5. For purposes of analysis, DR8, DR9, DR10, and DR blank were considered as a single group. A comparison of the *HLA-DR* gene frequencies in the 63 unrelated (independent) haplotypes found in 67 affected subjects, as compared with expectation based on a control population (Albert et al. 1984, p. 707), is significant (good-

Table 4

Probability, *P*, and χ'^2_m and *p* for HLA Allele Sharing IBD among LQT-affected Family Members, According to Indicated Criteria

	Initial QTC ≥0.47 or ≥0.45 with Symptoms[a]				Final Diagnoses[b]			
Family	No. affected	*P*	χ'^2_m	*p*	No. affected	*P*	χ'^2_m	*p*
CA001	7	.410	1.9932	.369	8	.367	2.2139	.331
CA002	4	.563	1.9617	.375	4	.563	1.9617	.375
CA003	3	1.000	.4153	.813	5	.938	.4937	.781
CA004	7	.270	3.1783	.204	7	.270	3.1783	.204
CA005	5	.141	5.0989	.078	6	.102	5.3096	.070
CA006	4	.438	2.7726	.250	3	.250	4.7726	.300
CA007	2	.250	4.7726	.092	3	.250	4.7726	.092
CA008	2	...	...	...	2	...	...	...
CA009	5	.563	1.4178	.492	5	.563	1.4178	.492
CA010	4	1.000	.6605	.719	4	1.000	.6605	.719
CA011	2	.250	4.7726	.092	2	.250	4.7726	.092
CA012	3	...	...	...	3	...	...	...
CA013	8	.272	3.3794	.185	7	.810	1.7635	.414
CA014	2	...	...	...	3	.250	4.7726	.092
CA015	5	.375	2.1357	.344	5	.375	2.1357	.344
Overall	63		32.5586	.114	67		38.2251	.058

Note.—*P* is probability of the observed or greater number of haplotype IBD matches; *p* values are based on χ^2 with 2 df for each family.

[a] Criteria used by Keating et al. (1991*a*, 1991*b*).

[b] See text.

ness-of-fit $\chi^2_7 = 18.54$, $p = .0098$). However, given the probable differences between the ethnic composition of the LQT syndrome sample (table 1) and that of the control population, we do not present the gene frequencies in the control population as any more than a general reference point. The frequency distribution of *HLA-DR* genes in affected subjects is also different from the frequency distribution in the combined group of unaffected and borderline subjects (internal controls) from the same LQT syndrome families (8 × 2 table; contingency $\chi^2_7 = 18.66$, $p = .0093$). Thus, there is a nonrandom distribution of *HLA-DR* genes in the 63 unrelated HLA haplotypes found in LQT syndrome patients, compared both with the 17 unrelated HLA haplotypes in internal controls and with approximate expectation based on Caucasian population gene frequencies.

If it is assumed that an effect of HLA region genes on LQT syndrome has been established by the global DR gene frequency analysis together with the PEDSCORE haplotype sharing analysis, the data in table 5 suggest that DR2 has a protective effect, and that DR7 has an enhancing effect, on the expression of the LQT phenotype. The one case in which DR2 was found in a haplotype that occurred in affected family members was in haplotype "b" in family CA013. In this, the only Spanish family, both parents and five (or six, in the initial diagnoses) of eight offspring had LQT syndrome (fig. 2), which suggests a strong genetic susceptibility. Haplotypes a, c, and d in this family were DR blank, DR5, and DR1 respectively. Haplotypes bearing DR7 were found in 25 of the 40 affected members of the nine families (CA001, CA002, CA003, CA005, CA008, CA009, CA010, CA012, and CA014) in which DR7 occurred. Of the 15 unrelated DR7 haplotypes, HLA-B57 was found in 4, B13 in 3, B35 and B44 in 2 each, and B8, B27, B51, and B62 in 1 each. Thus, the susceptibility conferred by *HLA*-region genes does not appear to be associated with any particular HLA-B allele. Among the five unrelated DR2 haplotypes, B62 was found in the one DR2 haplotype that occurred in the LQT syndrome patients (in CA013) and B7, B44, B52, and B62 were found in the four DR2 haplotypes that were found to occur in family members who were not (clearly) affected (table 5).

The DR7 association may be greater—and is certainly not less—in the subset of families in which linkage of H-*ras*-1 to *LQT* has been reported: 3 of 7 of the unrelated haplotypes in affected members of families CA005 and CA008 were DR7, whereas only 1 DR7 haplotype occurred among the 13 unrelated haplotypes in affected members of families CA004, CA007, and CA009. (Families in which the linkage relationship of H-*ras*-1 to *LQT* was unknown had 11 DR7 haplotypes among 43 unrelated haplotypes in affected family members.)

The MHC-haplotype sharing among affected siblings that was observed in these families and in the Japanese

Table 5

Frequency of HLA-DR Alleles in 63 Unrelated Haplotypes Found in 67 Affected Family Members, Compared with Unrelated Haplotypes Not Observed in Affected Family Members and in a General Caucasian Population

	Unrelated Haplotypes Found in Subjects (Final Diagnoses) Who Were						
	Affected[a]		Borderline[b]		Unaffected[c]		Control[d]
HLA-DR Gene	No.	%	No.	%	No.	%	%
1	5	7.9	2	25.0	0		9.4
2	1	1.6	1	12.5	3	33.3	16.2
3	9	14.3	0		0		11.4
4	9	14.3	1	12.5	1	11.1	13.3
5	12	19.0	3	37.5	3	33.3	14.6
6	8	12.7	1	12.5	1	11.1	11.4
7	15	23.8	0		0		12.3
8	1	1.6	0		1	11.1	2.9
9	1	1.6	0		0		0.8
10	0		0		0		0.8
Blank	2	3.2	0		0		7.0
Total	63	100.0	8	100.0	9	99.9	100.1

[a] QTc ≥.47 or ≥.45, with symptoms.
[b] Not "affected"; QTc = .45 or .46.
[c] Only individuals with QTc ≤.44.
[d] From Albert et al. (1984, p. 707).

family occurred for haplotypes inherited from the unaffected parent, as well as from the affected parent (figs. 1 and 2). We evaluated this observation with respect to the HLA-DR7–associated susceptibility. Four of the 25 affected persons with DR7 in the follow-up pedigrees (fig. 2; but with final diagnoses) were homozygous for DR7, inheriting DR7 from each parent. QT-interval data were not available for both parents of another 10 persons. Of the remaining seven individuals, three persons (one male and two female) in two different kindreds inherited DR7 from the affected parent, and eight persons (five male and three female) inherited DR7 from the unaffected parent in four kindreds (QTc of the parents = 0.41, 0.40, 0.45, and 0.39). The results suggest that *HLA*-region genes inherited from either parent may influence expression of the LQT phenotype. They are consistent with the prediction that *HLA*-region genes from the unaffected parent may make a contribution to disease susceptibility (Weitkamp 1981; Weitkamp et al. 1981).

Sex and HLA Genes

Females are more frequently affected with LQT syndrome than are males (Hashiba 1978, 1992; Moss et al. 1991). Among the subjects in the present report, 40/67 (59.7%) of the affected were female. The increased frequency in females occurred principally among persons who had QTc = 0.47–0.50. There was an equal frequency of males and females among the most severely affected, i.e., persons with symptoms or QTc ≥ 0.51 and among the borderline cases, i.e., persons without symptoms and with QTc = 0.45 or 0.46 (table 6; final diagnoses).

In order to evaluate a possible interactive effect between sex- and HLA-DR7–marked haplotypes in the expression of LQT syndrome, we tabulated the 133 HLA-DR–typed members of these pedigrees, according to the categories shown in table 6. When the twins were counted as one genetic event, DR7 occurred in 25/67 (37.3%) of affected family members and in 10/40 (25.0%) of unaffected siblings. Among these, the frequency of DR7 in affected females was 12/40 (30.0%), compared with 6/20 (30.0%) of unaffected female siblings. In contrast, the frequency of DR7 in affected males was 13/27 (48.1%), compared with 4/20 (20.0%) of unaffected male siblings (contingency $\chi^2_1 = 4.15$, $p = .042$).

Discussion

Model-independent methods of analysis of gene sharing among affected family members, such as used here, have gained increased attention in recent years (Weeks and Lange 1992; Brown et al. 1994). The disadvantage of affected-pedigree-member methods is that they are less powerful than lod-score methods, when a model can be correctly specified. However, the pedigree reported by Itoh et al. (1982), in which the apparent mode of transmission is dominant with complete penetrance (fig. 1), provides an unusual example to the contrary, because maximum IBD HLA haplotype sharing among affected family

Table 6

Sex of Family Members, According to Final Diagnosis, Mean QTc Interval, and Symptoms of LQT Syndrome

	No.[a]		
	Male	Female	% Female
Affected:			
QTc ≥.53	12 (4)	12 (4)	50.0
QTc = .51–.52	5 (3)	5 (0)	50.0
QTc = .49–.50	2 (0)	8 (4)	80.0
QTc = .46–.48[b]	7 (2)	14 (1)	66.7
QTc = .45–.46 (symptoms)	1 (1)	1 (1)	50.0
Overall	27 (10)	40 (10)	62.3
Borderline, QTc = .45–.46	9[c]	6	40.0
Unaffected, QTc ≤.44:			
Siblings of affected	9[d]	17	47.2
Other	5	10	66.7
Overall	24	27	52.9

[a] Numbers in parentheses are the number of persons with symptoms.
[b] At least one QTc ≥.47.
[c] Only eight males were typed for *HLA-DR*.
[d] Twins counted as one genetic event.

members occurred for haplotypes inherited from both parents of affected siblings. The additional information obtained by considering HLA haplotype transmission from the three unaffected parents to their affected offspring in the PEDSCORE analysis was greater than the additional information from the two unaffected offspring included in the lod-score analysis. Thus, PEDSCORE yielded an odds ratio of 1:2,048, whereas the odds ratio by standard linkage analysis was 1:512 (lod score = 2.71).

Elsewhere, we reported an analysis of HLA haplotype sharing in affected family members that gave a nonsignificant result (χ'^2_m = 30.03; 26 df; p = .27), considering only 52 of these family members as affected (Weitkamp et al. 1989). In that analysis, diagnosis was made by one of us (P.J.S.) on the basis of clinical history and clinical impression of the electrocardiographic patterns as well as on the basis of QT-interval length. In the current analyses of the same families, we initially scored 63 persons as affected by the (arbitrary) criteria of Keating et al. (1991*a*). Diagnoses were based on the earliest QTc in the database. Subsequently, 67 persons were scored as affected on the basis of all recorded QTc intervals. The reasons that QTc intervals from additional electrocardiograms were available in the database are undoubtedly varied, but they were unrelated to HLA status, which was unknown. There was apparently a bias toward retesting persons with a long QT interval. The initial characterization of clinical status has greater objectivity in the uniform assessment over all 134 subjects, but the second characterization may have some better diagnoses, on the basis of the additional data. Analysis of both sets of diagnoses fell short of providing statistical confirmation (p = .114 and .058) of HLA haplotype sharing among affected persons that was observed in the Japanese pedigree. On the basis of the additional independent evidence favoring an association between LQT syndrome and HLA-DR alleles, however, we conclude that an HLA effect on LQT syndrome cannot be ruled out.

Genetic complexity in the determination of the LQT phenotype is indicated by (1) the greater prevalence of the LQT syndrome or a longer QT interval in females than in males (Hashiba 1978, 1992; Moss et al. 1991; Vincent et al. 1992; present study), (2) an inability to define a clinical phenotype that is fully consistent with the Mendelian transmission patterns over several pedigrees (Weitkamp et al. 1989; present study) or that gives expected segregation ratios (Keating et al. 1991*a*), and (3) considerably lower postenrollment syncope and death rates among affected family members than among probands with LQT syndrome (Moss et al. 1991). The *LQT*-locus homogeneity that Keating et al. (1991*b*) proposed has not been confirmed in follow-up reports of families in which a major LQT syndrome locus is not linked to H-*ras*-1 (Kerem et al. 1992; Towbin et al. 1992; Benhorin et al. 1993; Curran et al. 1993). More than locus heterogeneity, however, we propose that the genetic determination of susceptibility to the LQT syndrome is complex, with at least one locus in the *HLA* region modifying or modulating the expression of the phenotype. Gender may also have a role. The analysis by Vincent et al. (1992) supports this concept. On the assumption that H-*ras*-1 alleles could be used to identify unambiguously carriers and noncarriers of a primary *LQT* gene in the Utah kindred, they noted that diagnosis of

LQT syndrome on the basis of QTc intervals lacked complete sensitivity or specificity in the 0.42–0.46 range for males and the 0.45–0.47 range for females. As further evidence of complexity, we have noted in this study not only "incomplete penetrance" (arguably, incomplete sensitivity of the QTc interval as an indicator of phenotype), but also the fact that spouses of an affected parent of affected offspring often had a QTc of 0.45 or 0.46—in the ambiguous range. *HLA*-region genes may be one specific element affecting expression of the LQT phenotype. HLA-DR2 had a protective effect, and, overall, HLA-DR7 may increase vulnerability to the LQT syndrome, as defined by a lengthened QT interval. The frequency of DR7 was higher in affected males (48%) than in unaffected male siblings (20%), but for females, the frequency was the same for affected and unaffected siblings (both 30%).

HLA-DR alleles have been associated with a number of disorders, mostly autoimmune diseases with a relatively low risk for relatives of the proband. Many of these diseases are more common in females than in males, and, in the case of multiple sclerosis, an interactive effect between sex and *HLA* genes has been proposed (Weitkamp 1983). Of particular interest as a model for the genetic basis of LQT syndrome is insulin-dependent diabetes mellitus (IDDM). The major histocompatibility complex (MHC) component of susceptibility to this well-studied disorder has been difficult to define, because of the extreme linkage disequilibrium in the *HLA* region. However, one component of susceptibility in White populations results from different combinations of the *cis*- and *trans*-encoded α and β heterodimers of the HLA-DQ molecule (Kwok et al. 1990; Ronningen et al. 1991; Khalil et al. 1992). DR molecules additionally influence susceptibility (Sheehy et al. 1989), as do in fact MHC genes outside the class II region (Christiansen et al. 1990; Degli-Esposti et al. 1992; Tienari et al. 1992). HLA-DR2 has been associated with a protective effect. The 5′ insulin-gene polymorphism on chromosome 11p15 (near the H-*ras*-1 gene on 11p15) marks a second region determining susceptibility to IDDM (Raffel et al. 1992). In the nonobese diabetic mouse model of IDDM, genes at several unlinked loci interact with MHC genes to produce susceptibility (Cornall et al. 1991; Garchon et al. 1991; Todd et al. 1991). We do not have sufficient data to determine whether the HLA effect in LQT syndrome is limited to the subset of families that shows linkage between LQT and the H-*ras*-1 gene or to the subset of families that does not show such linkage. However, in both the affected-pedigree-member HLA haplotype IBD matching data and the HLA-DR association data there is somewhat greater evidence for an effect in the two H-*ras*-1–linked families (CA005 and CA008) than in the three families (CA004, CA007, and CA009) not linked to H-*ras*-1.

Although the familial risk for LQT syndrome is high, it is not a strictly dominant disorder. The reports, in some families, of linkage of a locus for the LQT syndrome to the H-*ras*-1 locus on chromosome 11p15, as well as the probable DR2 protective effect reported here, are intriguing in light of similar observations in the IDDM data. Indeed, QT-interval lengthening is a feature of diabetic autonomic neuropathy (Ewing et al. 1991). Keating et al. (1991*a*) assumed a penetrance of 90% in the analysis that yielded a lod score of 16.44 for linkage of H-*ras*-1 to the hypothesized *LQT* locus with no recombinants. Is it possible that the "linkage" of a locus on chromosome 11p15 to the LQT syndrome locus detected by lod-score analysis is simply an effect that genes on chromosome 11 may have on the expression of the LQT syndrome? In this regard, the exclusion of linkage between *LQT* and the H-*ras*-1 locus up to 6 cM (multipoint lod score <-2.0) in each of two pedigrees (K1756 and K1977) reported by the same authors (Curran et al. 1993) is of interest. Both pedigrees show some 11p15.5 gene sharing among affected family members (from the unaffected parent). The data in these two pedigrees are insufficient to determine whether the model of linkage between Mendelian loci does not efficiently detect the effect of 11p15.5 genes (from either parent) in producing the *LQT* phenotype; unfortunately, the large pedigree reported by Keating et al. (1991*a*) cannot be evaluated, because the pedigree structure was altered to protect confidentiality.

The model that emerges for IDDM is one in which differing combinations of class II and non–class II MHC genes interact with genes at non-MHC loci to produce the susceptibility that permits, with differing age at clinical onset (Collat-Zucman et al. 1992), the immune destruction of pancreatic beta cells. We contemplate a similar model for LQT syndrome: one in which genes on chromosome 6, 11, and possibly other chromosomes interact variously with sex to produce a longer QT interval and, in some cases, an LQT syndrome of varying severity. There is evidence for impairment of transmembrane signaling in potassium ion channels in the LQT syndrome (for a discussion of the molecular physiology, see Vincent 1992). This electrocardiographic repolarization abnormality offers promise for the investigation of the genetic and pathophysiological basis of these effects.

Acknowledgments

We thank Patricia Caldwell, Patricia Ames, and Mary True for assistance in manuscript preparation. This project was supported in part by grant HL-33843 from the National Institutes of Health.

References

Albert ED, Baur MP, Mayr WR (eds) (1984) Histocompatibility testing 1984. Springer, Berlin

Alper CA, Boenisch T, Watson L (1972) Genetic polymorphism

in human glycine-rich beta-glycoprotein. J Exp Med 135:68–80

Bazett HC (1920) An analysis of the time-relations of electrocardiograms. Heart 7:353–370

Benhorin J, Kalman YM, Medina A, Towbin J, Rave-Harel N, Dyer TD, Blangero J, et al (1993) Evidence of genetic heterogeneity in the long QT syndrome. Science 260:1960–1962

Brown DL, Gorin MB, Weeks DE (1994) Efficient strategies for genomic searching using the affected-pedigree-member method of linkage analysis. Am J Hum Genet 54:544–552

Bruning JW (1976) HL-A typing with the use of fresh or frozen fluorescein-labelled lymphocytes. In: Ray JG, Hare DB, Pedersen PD, Mullaly DI (eds) NIAID manual of tissue typing techniques 1976–1977. Government Printing Office, Washington, DC, pp 29–30

Christiansen FT, Sauracker GC, Leaver AL, Tokunaga K, Cameron PU, Dawkins RL (1990) Characterization of MHC ancestral haplotypes associated with insulin-dependent diabetes mellitus: evidence for involvement of non-HLA genes. J Immunogenet 17:379–386

Collat-Zucman S, Garchon H-J, Timsit J, Assan R, Boitard C, Djilali-Saiah I, Bougneres P, et al (1992) Age-dependent HLA genetic heterogeneity of type 1 insulin-dependent diabetes mellitus. J Clin Invest 90:2242–2250

Cornall RJ, Prins J-B, Todd JA, Pressey A, DeLarato N, Wicker LS, Peterson LB (1991) Type 1 diabetes in mice is linked to the interleukin-1 receptor and Lsh/Ity/Bcg genes on chromosome 1. Nature 353:262–265

Curran M, Atkinson D, Timothy K, Vincent GM, Moss AJ, Leppert M, Keating M (1993) Locus heterogeneity of autosomal dominant long QT syndrome. J Clin Invest 92:799–803

Darke C (1976) Ethidium bromide microtoxicity test. In: Ray JG, Hare DB, Pedersen PD, Mullaly DI (eds) NIAID manual of tissue typing techniques 1976–1977. Government Printing Office, Washington, DC, pp 35–36

Degli-Esposti MA, Abraham LJ, McCann V, Spies T, Christiansen FT, Dawkins RL (1992) Ancestral haplotypes reveal the role of the central MHC in the immunogenetics of IDDM. Immunogenetics 36:345–356

Ewing DJ, Boland O, Neilson JMM, Cho DG, Clarke BF (1991) Autonomic neuropathy, QT interval lengthening, and unexpected deaths in male diabetic patients. Diabetologia 34:182–185

Garchon H-J, Bedossa P, Eloy L, Bach J-F (1991) Identification and mapping to chromosome 1 of a susceptibility locus for periinsulitis in non-obese diabetic mice. Nature 353:260–262

Giuffre RM, Hejtmancik JF, McCabe ERB, Towbin JA (1990) Long QT (Romano-Ward) syndrome: molecular genetic evidence against tight HLA linkage. Am J Hum Genet Suppl 47: A180

Hashiba K (1978) Hereditary QT prolongation syndrome in Japan: genetic analysis and pathologic findings of the conducting system. Jpn Circ J 42:1133–1150

——— (1992) Sex differences in phenotypic manifestation and gene transmission in the Romano-Ward syndrome. Ann NY Acad Sci 644:142–156

Itoh S, Mumemura S, Satoh H (1982) A study of the inheritance pattern of Romano-Ward syndrome. Clin Pediatr (Phila) 21: 20–24

Keating M, Atkinson D, Dunn C, Timothy K, Vincent GM, Leppert M (1991*a*) Linkage of a cardiac arrhythmia, the long QT syndrome, and the Harvey *ras*-1 gene. Science 252:704–706

Keating M, Dunn C, Atkinson D, Timothy K, Vincent GM, Leppert M (1991*b*) Consistent linkage of the long-QT syndrome to the Harvey *Ras*-1 locus on chromosome 11. Am J Hum Genet 49:1335–1339

Kerem B, Benhorin J, Kalman YM, Medina A, Dyer TD, Blangero J, MacCluer JW (1992) Evidence for genetic heterogeneity in the long QT syndrome. Am J Hum Genet Suppl 51:A192

Khalil I, Deschamps I, Lepage V, Al-Daccak R, Degos L, Hors J (1992) Dose effect of cis- and trans-encoded HLA-DQαβ heterodimers in IDDM susceptibility. Diabetes 41:378–384

Kwok WW, Mickelson E, Masewicz S, Milner ECB, Hansen J, Nepom GT (1990) Polymorphic DQα and DQβ interactions dictate HLA class II determinants of allo-recognition. J Exp Med 171:85–95

Lamm LU, Weitkamp LR, Jensen O, Brunn Pedersen G, Kissmeyer-Nielsen F (1978) On the mapping of PGM_3, GLO, and HLA. Tissue Antigens 11:132–138

Lancaster HO (1949) The combination of probabilities arising from data in discrete distributions. Biometrika 36:370–382

McKusick VA (ed) Mendelian inheritance in man, 10th ed. Johns Hopkins University Press, Baltimore

Mamelka PM, Dyke B, MacCluer JW (1990) Pedigree/Draw. PGL tech rep 1, Population Genetics Laboratory, Department of Genetics, Southwest Foundation for Biomedical Research, San Antonio

Melki J, Kaplan J, Luce V, Halley L, Clemenceau S, Kaplan C, Baule MS, et al (1987) Long QT (Romano-Ward) syndrome: further definition of the linkage to the HLA loci. Cytogenet Cell Genet 46:661

Moss AJ (1986) Prolonged QT syndrome. JAMA 256:2985–2987

Moss AJ, Schwartz PJ, Crampton RS, Tzivoni D, Locati EH, MacCluer J, Hall WJ, et al (1991) The long QT syndrome: prospective longitudinal study of 328 families. Circulation 84: 1136–1144

Raffel LJ, Hitman GA, Toyoda H, Karam JH, Bell GI, Rotter JI (1992) The aggregation of the 5′ insulin gene polymorphism in insulin dependent (Type I) diabetes mellitus families. J Med Genet 29:447–450

Romano C (1965) Congenital cardiac arrhythmia. Lancet 1:658–659

Romano C, Gemme G, Pongiglione R (1963) Aritmie cardiache rare dell'eta pediatrica. Clin Pediatr (Phila) 45:656–683

Ronningen KS, Spurkland A, Iwe T, Vartdal E, Thorsby E (1991) Distribution of HLA-DRB1, -DQA1, and -DQB1 alleles and DQA1-DQB1 genotypes among Norwegian patients with insulin-dependent diabetes mellitus. Tissue Antigens 37:105–111

Schwartz PJ, Periti M, Malliani A (1975) The long QT syndrome. Am Heart J 89:378–390

Sheehy MJ, Scharf SJ, Rowe JR, Neme de Gimenez MH, Mesky LM, Erlich HA, Nepom BS (1989) A diabetes susceptibility haplotype is best defined by a combination of HLA-DR and DQ alleles. J Clin Invest 83:830–835

Tienari PJ, Tuomilehto-Wolf E, Tuomilehto J, Peltonen L, the DIME Study Group (1992) HLA haplotypes in type 1 (insulin-dependent) diabetes mellitus: molecular analysis of the HLA-DQ locus. Diabetologia 35:254–260

Todd JA, Hitman TJ, Cornall RJ, Ghosh S, Hall JRS, Hearne M,

Knight AM, et al (1991) Genetic analysis of autoimmune type 1 diabetes mellitus in mice. Nature 351:542–547

Towbin JA, Pagotto L, Siu B, Robinson J, Moss A, McCabe ERB, Hejtmancik JF (1992) Romano-Ward long QT syndrome (RWLQTS): evidence of genetic heterogeneity. Pediatr Res 31:23a

Vincent GM (1992) Hypothesis for the molecular physiology of the Romano-Ward Long QT syndrome. J Am Coll Cardiol 20: 500–503

Vincent GM, Timothy KW, Leppert M, Keating M (1992) The spectrum of symptoms and QT intervals in carriers of the gene for the Long-QT syndrome. N Engl J Med 327:846–852

Ward OC (1964) A new familial cardiac syndrome in children. J Ir Med Assoc 54:103–106

Weeks DE, Lange K (1992) A multilocus extension of the affected-pedigree-member method of linkage analysis. Am J Hum Genet 50:859–868

Weitkamp LR (1981) HLA and disease: Predictions for *HLA* haplotype sharing in families. Am J Hum Genet 33:776–784

——— (1983) Multiple sclerosis susceptibility: interaction between sex and HLA. Arch Neurol 40:399–401

Weitkamp LR, Lewis RA (1992) PEDSCORE analysis of identical by descent (IBD) marker allele distributions among family members with cutaneous melanoma. Cytogenet Cell Genet 59: 231–233

Weitkamp LR, Moss AJ (1985) Romano-Ward (long QT) syndrome locus is probably linked to the HLA loci. Cytogenet Cell Genet 40:775

Weitkamp LR, Moss AJ, Schwartz PJ, Locati E, Tzivoni D, Vincent GM, Robinson J, et al (1989) Analysis of HLA haplotypes in long QT syndrome: withdrawal of the preliminary assignment of LQT to the HLA linkage group. Cytogenet Cell Genet 51:1106

Weitkamp LR, Stancer HC, Persad E, Flood C, Guttormsen S (1981) Depressive disorders and HLA: a gene on chromosome 6 that can affect behavior. N Engl J Med 305:1301–1306