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Chromosome 14 and Late-Onset Familial Alzheimer Disease (FAD)

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Summary

Familial Alzheimer disease (FAD) is genetically heterogeneous. Two loci responsible for early-onset FAD have been identified: the amyloid precursor protein gene on chromosome 21 and the as-yet-unidentified locus on chromosome 14. The genetics of late-onset FAD is unresolved. Maximum-likelihood, affected-pedigree-member (APM), and sib-pair analyses were used, in 49 families with a mean age at onset ≥60 years, to determine whether the chromosome 14 locus is responsible for late-onset FAD. The markers used were D14S53, D14S43, and D14S52. The LOD score method was used to test for linkage of late-onset FAD to the chromosome 14 markers, under three different models: age-dependent penetrance, an affected-only analysis, and age-dependent penetrance with allowance for possible age-dependent sporadic cases. No evidence for linkage was obtained under any of these conditions for the late-onset kindreds, and strong evidence against linkage (LOD score ≤ −2.0) to this region was obtained. Heterogeneity tests of the LOD score results for the combined group of families (early onset, Volga Germans, and late onset) favored the hypothesis of linkage to chromosome 14 with genetic heterogeneity. The positive results are primarily from early-onset families. APM analysis gave significant evidence for linkage of D14S43 and D14S52 to FAD in early-onset kindreds ($P < .02$). No evidence for linkage was found for the entire late-onset family group. Significant evidence for linkage to D14S52, however, was found for a subgroup of families of intermediate age at onset (mean age at onset ≥60 years and <70 years). These results indicate that the chromosome 14 locus is not responsible for Alzheimer disease in most late-onset FAD kindreds but could play a role in a subset of these kindreds.

Introduction

Early-onset familial Alzheimer disease (FAD) is genetically heterogeneous. In a subset of early-onset families, mutations in the amyloid precursor protein (APP) gene on chromosome 21 cause the disease (Chartier-Harlin et al. 1991; Goate et al. 1991; Murrell et al. 1991; Mullan et al. 1992*a*). In a larger group of early-onset kindreds, an as-yet-unidentified locus on chromosome 14 is responsible for Alzheimer disease (AD) (Schellenberg et al. 1992*a*). For both the chromosome 14 locus and the APP gene, inheritance is apparently autosomal dominant.

The genetics of late-onset FAD has not been resolved. (Late-onset FAD kindreds are arbitrarily defined as those having a mean age at onset ≥60 years.) Evidence from epidemiologic case-control surveys (Mohs et al. 1987; van Duijn et al. 1991), recent twin studies (Rappaport et al. 1991; Bergem et al. 1992; Breitner et al. 1992), and preliminary studies of nuclear

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Table 1

Characteristics of FAD Families

Family	No. Affected[a]	No. Autopsied	No. of Affecteds Sampled[a]	No. of Subjects Sampled	Mean Age at Onset ± SD (*n*) Range (years)	Probability of Linkage to D14S43		
						Model 1[b]	Model 2	Model 3
L	16	9[c]	7	23	42 ± 4.6 (16) 30–48	1.00 (1.00)	1.00	.97
AM	8	2[d]	5	10	42 ± 3.2 (8) 36–46	.20 (.65)	.31	.60
HR-XV	12	1[d]	4	15	42 ± 3.9 (6) 37–47	.03 (.34)	.05	.71
KG	5	3	1	11	44 ± 1.0 (5) 43–45	.65 (.93)	.77	.72
V	8	1	2	9	46 ± 3.8 (7) 41–50	.02 (.20)	.04	.54
HR-I	13	6	3	31	47 ± 4.6 (12) 40–55	.87 (.97)	.93	.91
HR-XIII	4	4	0	11	47 ± 3.6 (4) 41–51	.33 (.77)	.47	.57
LH(603)	23	6	5	27	48 ± 6.5 (18) 37–68	.84 (.97)	.90	.90
HR-XVII	7	0	2	12	51 ± 2.9 (7) 45–55	nd	nd	nd
R[e]	23	4	4	30	51 ± 7.1 (17) 40–67	.26 (.70)	.39	.53
SNW	19	5	4	17	52 ± 2.5 (7) 48–56	.97 (1.00)	.99	.70
W[e]	4	1	2	4	54 ± 3.9 (4) 48–58	.03 (.28)	.05	.65
HD[e]	23	1	6	14	59 ± 10.4 (18) 46–82	.01 (.61)	.03	.70
HB[e]	21	4[f]	4	24	60 ± 7.2 (20) 47–75	.00 (.00)	.00	.70
SB	5	1	2	5	62 ± 7.3 (5) 55–75	nd	nd	nd
WFL[e]	6	2	2	15	64 ± 7.6 (6) 55–76	.11	.17	.56
HR-XI	5	2	3	10	64 ± 5.4 (5) 56–69	.17	.28	.55
MMM[g]	5	1	1	3	64 ± 0.5 (3) 64–65	.20	.29	.54
BEH	7	2	2	2	65 ± 6.7 (3) 58–74	.15	.32	.49
CK	3	3	2	3	65 ± 6.8 (3) 56–72	.14	.24	.49
KS[e]	12	3	6	24	65 ± 5.1 (11) 55–71	.01 (.53)	.01	.57
BL	6 (1)	1	4	9	66 ± 4.3 (5) 58–70	.06	.18	.54
ORGK	6 (1)	2	3	7	66 ± 8.3 (5) 55–81	.00	.09	.33
ORGL	3	1	2	2	66 ± 2.5 (2) 63–68	.30	.31	.63
BOB	5 (2)	2	2	5	67 ± 9.3 (5) 56–85	.14	.22	.65
CFD	5 (1)	1	3	5	67 ± 8.2 (3) 56–75	.06	.31	.34
HTB	6 (5)	1[h]	4	8	67 ± 3.9 (5) 61–80	.00	.08	.26
ORWW	8	1	3	4	67 ± 7.0 (4) 60–78	.10	.48	.49
P	7	1	3	4	67 ± 4.4 (4) 62–74	.03	.20	.38
HR-XII	8 (1)	2	4	11	68 ± 6.0 (8) 57–74	.01	.02	.37
LQ	8	2	3	5	68 ± 4.6 (8) 59–75	.40	.44	.69
ORLR	4	1	2	2	68 ± 0.5 (3) 68–69	.27	.39	.61
ORMK	2	0	2	5	68 ± 1.5 (2) 66–69	.10	.30	.46
PH	5 (3)	2	5	13	68 ± 10 (5) 51–80	.20	.31	.57
WLA	3 (1)	1	3	3	68 ± 4.0 (3) 62–71	.18	.30	.52
BCR	10 (1)	1	5	6	69 ± 6.2 (9) 63–80	.01	.26	.37
ORHW	3	0	1	2	69 ± 8.2 (3) 60–80	.19	.31	.54
ORPH	7	1	2	7	69 ± 8.1 (4) 59–81	.29	.40	.59
RR[i]	4 (2)	1	2	8	69 ± 4.0 (4) 65–80	.16	.30	.58
CSF	4	2	2	7	70 ± 5.3 (4) 64–77	.02	.25	.32
FZG[g]	4 (1)	1	2	7	70 ± 4.9 (4) 62–74	.14	.30	.50
MI	4 (1)	1	2	3	70 ± 4.0 (4) 65–75	.10	.30	.48
ORAS	5	1	2	3	70 ± 1.5 (2) 68–71	.05	.12	.50
KAM	7 (5)	0	5 (1)	10	71 ± 7.8 (6) 57–81	.17	.37	.57
ORDF	3	0	2	2	71 ± 0.9 (3) 70–72	.12	.25	.46
FH	4	1	2	4	72 ± 7.8 (4) 64–81	.25	.30	.51
HR-IX	7	3	5	11	72 ± 1.9 (6) 69–75	.01	.15	.45
ORCW	7	1	5	7	72 ± 7.3 (7) 59–82	.00	.09	.52
ORRH	3	0	2	3	72 ± 6.0 (2) 66–78	.31	.33	.54
OREB	5 (1)	0	2 (1)	3	73 ± 0.5 (2) 72–74	.00	.25	.32

(*continued*)

Table 1 (continued)

Family	No. Affected[a]	No. Autopsied	No. of Affecteds Sampled[a]	No. of Subjects Sampled	Mean Age at Onset ± SD (*n*) Range (years)	Probability of Linkage to D14S43		
						Model 1[b]	Model 2	Model 3
FF	4 (2)	0	4	5	74 ± 7.1 (4) 64–84	.13	.27	.48
LJT	6	1	4	8	74 ± 6.5 (4) 65–83	.00	.03	.31
ORJM	5	2	0	2	74 ± 4.3 (3) 70–80	.20	.31	.54
ORLA	5	2	3	3	74 ± 1.2 (5) 72–75	.18	.28	.52
ORLH	7	1	4	5	75 ± 1.5 (5) 72–76	.12	.20	.48
ORSK	8	1	3	4	75 ± 0.0 (2) 75	.52	.41	.71
QDH	4	2	2	3	75 ± 2.1 (3) 72–77	.34	.40	.62
ORJV	6	1	3	3	77 ± 2.5 (3) 74–80	.02	.43	.32
JR	3	2	1	5	78 ± 1.9 (3) 75–79	.18	.30	.51
ORHM	5	0	3	5	78 ± 5.4 (3) 74–86	.26	.31	.65
ORKD	3	0	2	6	78 ± 1.4 (3) 76–79	.27	.38	.54
ORPK	7	1	2	4	78 ± 2.9 (3) 75–82	.01	.34	.34
ORSS	5	0	2	6	79 ± 4.5 (4) 71–83	.26	.31	.59
EL	4	1	3	3	80 ± 3.7 (4) 75–85	.01	.31	.32
Total	447 (28)	105	185	516				

NOTE.—In comparison with previous work (Schellenberg et al. 1992*a*), additional subjects have been genotyped for the HR-XV, HR-I, R, and HD families. nd = not determined.

[a] Values in parentheses indicate number of family members with the diagnosis of possible AD or demented, by family history.

[b] Values in parentheses are probabilities of linkage when only the VG and ENVG families are considered. Probabilities were determined using the A-test.

[c] Includes one unaffected subject autopsied.

[d] Brain biopsy.

[e] Volga German.

[f] Includes two unaffected subjects autopsied.

[g] Black Sea German.

[h] Autopsy pending.

[i] German from Russia (Russian origin unknown).

families with two affected parents (Bird and Nemens 1992) indicate that defective genes play a role in late-onset disease. However, the mode of inheritance is not clear; both autosomal dominant inheritance (Mohs et al. 1987) and multifactorial models have been proposed (Farrer et al. 1991). Also, nongenetic factors may cause some late-onset AD. Genetic analysis of late-onset FAD is problematic in part because AD is a common disease in the elderly. Potentially confounding factors include age censoring, the potential mixture of sporadic and genetic cases in families ascertained for the presence of multiple cases, multiple sources of the disease gene in a single pedigree, the difficulty in obtaining large pedigrees, and uncertainty as to the mode of inheritance.

The location of gene(s) contributing to late-onset FAD has not been established. No APP mutations which cause late-onset FAD have been identified. Also, for late-onset families, linkage analysis for chromosome 21 markers is consistently negative under the assumption of single-locus inheritance (Pericak-Vance et al. 1988; Schellenberg et al. 1988, 1991; Kamino et al. 1992). A late-onset locus on chromosome 19 has been suggested (Schellenberg et al. 1987, 1992*b*; Pericak-Vance et al. 1991; Strittmater et al. 1993). In light of the recent identification of an early-onset chromosome 14 FAD locus, we used chromosome 14 markers to examine the role of the chromosome 14 FAD locus in a large group of late-onset FAD families.

Subjects, Material, and Methods

FAD Families

Families (table 1) were evaluated either by the University of Washington Alzheimer's Disease Research Center, the Oregon Health Sciences University Alzheimer's Disease Center, or the University of Minnesota AD research group. Affected subjects were evalu-

Table 2

Two-Point LOD Scores for Linkage of Late-Onset AD to Chromosome 14 Markers

Marker[a] and Model	Z at θ = .001	.05	.10	.15	.20	.30	.40
D14S52:							
1	−6.10 (−13.82)	−2.83 (−6.07)	−1.53 (−3.54)	−.79 (−2.08)	−.35 (−1.17)	.00 (−.28)	.03 (−.04)
2	−1.99 (−4.38)	−.45 (−1.66)	.08 (−.61)	.29 (−.07)	.35 (.17)	.25 (.24)	.07 (.08)
3	−4.06 (−5.91)	−2.71 (−3.84)	−1.85 (−2.54)	−1.26 (−1.68)	−.83 (−1.09)	−.33 (−.40)	−.07 (−.08)
D14S43:							
1	−19.59 (−24.28)	−8.43 (−10.00)	−4.97 (−5.73)	−3.03 (−3.36)	−1.82 (−1.92)	−.57 (−.49)	−.08 (−.07)
2	−10.36 (−12.59)	−5.30 (−6.74)	−3.12 (−4.09)	−1.88 (−2.52)	−1.11 (−1.52)	−.33 (−.47)	−.03 (−.09)
3	−5.36 (−6.09)	−3.15 (−3.52)	−2.01 (−2.18)	−1.30 (−1.35)	−.82 (−.82)	−.27 (−.25)	−.04 (−.05)
D14S53:							
1	−10.14 (−11.72)	−4.17 (−4.78)	−2.07 (−2.45)	−.94 (−1.18)	−.31 (−.45)	.15 (.12)	.12 (.12)
2	−6.47 (−6.98)	−2.79 (−2.98)	−1.32 (−1.36)	−.56 (−.54)	−.15 (−.11)	.12 (.16)	.09 (.10)
3	−3.47 (−3.73)	−1.94 (−2.18)	−1.14 (−1.36)	−.66 (−.84)	−.34 (−.49)	−.05 (−.12)	.02 (.00)

NOTE.—LOD scores not in parentheses are for autopsy-documented families, and values in parentheses are for autopsied and nonautopsied pedigrees combined.

[a] The genetic relationship of these markers on the long arm of chromosome 14 is centromere–D14S52–(20.7 cM)–D14S43–(2.5 cM)–D14S53–telomere (Schellenberg et al. 1992*a*; Wang and Weber 1992).

ated, the diagnosis of AD assigned, and autopsies performed, as described elsewhere (McKhann et al. 1984; Bird et al. 1988, 1989; Schellenberg et al. 1991). Autopsy documentation of AD was available for 40 of the 49 families studied.

Linkage Analysis

Dinucleotide repeat polymorphism genotypes were determined as described elsewhere (Schellenberg et al. 1992*b*). LOD scores were calculated using the computer program LIPED modified to handle up to 20 alleles and with the capacity to handle different alleles for each family. LIPED was also modified to incorporate a cumulative normal sporadic AD age-at-onset function (model 3 below) by using the approach suggested by Margaritte et al. (1992). LOD scores were computed using three different models, each under the assumption of autosomal dominant inheritance. Model 1 assumed age-dependent penetrance with a cumulative normal age-at-onset correction (Schellenberg et al. 1991). The age curve was constructed using family-specific means and an overall SD of 7.18 years for late-onset families, 8.62 years for Volga German (VG) families, and 5.60 years for early-onset non-Volga German (ENVG) families. For model 2, LOD scores were calculated by setting the penetrance of the AD genotype to a fixed value of 1% (also referred to as "affected-only analysis"), which effectively ignores the AD phenotypes of the unaffected, but at-risk, subjects but which uses their marker genotypes in the likelihood computations. Model 3 was the same as model 1, with the addition of a cumulative normal sporadic age-at-onset function with a mean of 110 years and an SD of 14 years. This model effectively corrects for sporadic AD patients in the families by altering the probability that an affected individual is a sporadic versus an FAD gene carrier; as the age at onset of a subject increases, the probability that the subject is a sporadic subject increases (Magaritte et al. 1992). The probability that FAD in a given family is the result of the chromosome 14 locus was taken from the within-group A-test for heterogeneity (Smith 1961; Hodge et al. 1983).

Affected-Pedigree-Member (APM) Method

Families in which genotypes from two or more affected subjects could either be directly typed or completely inferred were analyzed by the APM method of Weeks and Lange (1988) using a computer program provided by D. Weeks. The APM method tests the null hypothesis of independent segregation of the disease with marker alleles. The test statistic approximates the normal distribution; a one-sided test is used to determine the significance of the results. The test statistic was computed for three weighting schemes: $f(p) = 1$, $f(p) = 1/\sqrt{p}$, and $f(p) = 1/p$, where p is the allele frequency of the test marker. The first scheme gives no

Table 3

Two-Point LOD Scores for Linkage of D14S43 to Late-Onset AD, Divided by Age at Onset

Mean Age and Model	Z at θ = .001	.05	.10	.15	.20	.30	.40
Family mean age at onset >60 and <70 years:							
1	−10.43 (−10.81)	−4.66 (−4.99)	−2.87 (−3.13)	−1.82 (−2.03)	−1.15 (−1.31)	−.41 (−.48)	−.08 (−.10)
2	−4.57 (−4.83)	−2.45 (−2.64)	−1.45 (−1.59)	−.86 (−.96)	−.50 (−.57)	−.14 (−.17)	−.01 (−.02)
3	−3.40 (−3.42)	−1.97 (−1.98)	−1.27 (−1.28)	−.84 (−.84)	−.54 (−.55)	−.20 (−.20)	−.04 (−.04)
Family mean age at onset ≥70 years:							
1	−11.97 (−13.80)	−4.98 (−5.13)	−2.88 (−2.69)	−1.73 (−1.41)	−1.02 (−.67)	−.30 (−.04)	−.03 (.06)
2	−6.53 (−8.05)	−3.54 (−4.34)	−2.20 (−2.70)	−1.39 (−1.72)	−.87 (−1.08)	−.29 (−.36)	−.04 (−.06)
3	−2.66 (−2.67)	−1.63 (−1.54)	−1.05 (−.90)	−.67 (−.51)	−.42 (−.26)	−.14 (−.03)	−.02 (.02)

NOTE.—LOD scores not in parentheses are for autopsy-documented families, and values in parentheses are for autopsied and nonautopsied pedigrees combined.

weight to allele frequencies, whereas the second and third schemes make the sharing of a rare allele a more significant event than the sharing of a common allele. The third scheme, however, usually leads to a non-normality of the test statistics. The intermediate function is therefore a good compromise for incorporating an allele frequency function and generating a normal distribution. When the null hypothesis was rejected (i.e., test statistics >1.645; theoretical P value < .05), the segregation of the marker alleles through the pedigrees was simulated to examine the validity of the normality approximation; 5,000 iterations were performed, and the mean, variance, and upper 95th and 99th percentiles were determined empirically.

Affected-Sib-Pair (ASP) Method

The ASP method tests the distribution of marker-haplotype sharing among affected siblings for evidence of linkage (Penrose 1935; Motro and Thomson 1985). Like the APM method, the ASP analysis uses only affected individuals and is independent of the mode of inheritance. The major distinction between ASP and APM is that APM is based on identity by state, whereas ASP is based on identity by descent; thus the ASP method is not dependent on allele frequency estimates.

For each nuclear family, the parental haplotypes were constructed using the D14S43 and D14S53 genotypes. These markers were chosen because D14S43 and D14S53 are closely linked (separated by 2.5 cM), while D14S52 is more distant, being 20.7 and 23.2 cM from D14S43 and D14S53, respectively (Schellenberg et al. 1992*a*). For families in which the four parental haplotypes could be distinguished, we determined whether each ASP shared 2, 1, or 0 haplotypes. The observed haplotype-sharing distribution was compared with the χ^2 test to the expectations under the null hypothesis of no linkage. The issue of nonindependence in the families with more than two affected sibs was addressed, as suggested by Motro and Thomson (1985).

Results

Highly significant positive LOD scores were obtained when ENVG kindreds were tested for linkage to the chromosome 14 markers D14S43, D14S53, and D14S52 (Schellenberg et al. 1992*a*). Age-at-onset means for this group had a range of 42–51 years. In contrast, for the VG families, which have an intermediate age at onset (group mean = 57 years, and family means have a range of 52–65 years), significant evidence excluding linkage to these markers was obtained (Schellenberg et al. 1992*a*). The VG kindreds are considered separately, since this group of families is probably the result of a common genetic founder (Bird et al. 1988).

We tested for linkage of D14S52, D14S53, and D14S43 to FAD in 49 late-onset kindreds (table 1) by using the three models described in Subjects, Material, and Methods. The summed LOD scores for the late-onset group were negative under all three models (table 2). When only autopsy-documented families (40 kindreds) were tested, linkage to D14S43 was formally excluded to a recombination fraction (θ) from approximately .10 to .15 (LOD score [Z] ≤ -2.0), depending on

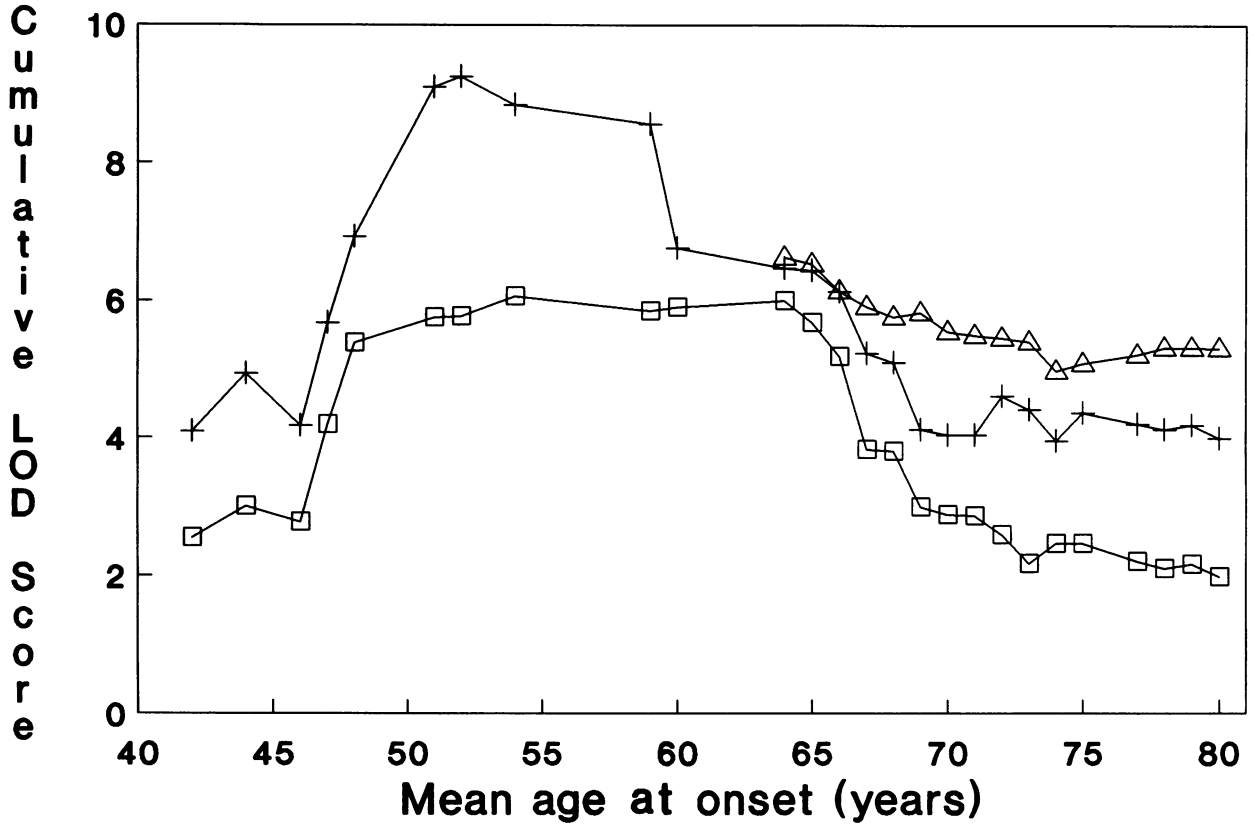

Figure 1 Effect of age on the cumulative LOD score for linkage of D14S43 to FAD, using three different genetic models. LOD scores were summed for all families with a mean age at onset less than or equal to the age given. Plus signs (+) denote model 1; squares denote model 2; and triangles denote model 3. LOD scores given are Z_{max} for a given age group; the values of θ vary from .001 to .20.

the model used. Similar results were obtained when non-autopsy-documented families were included. When families were further subdivided into one group with a mean age at onset between 60 and 69 years and a second group with a mean age at onset >70 years, again no evidence for linkage in either group under any of the models tested was obtained (table 3). When the cumulative LOD scores for D14S43 were plotted versus age at onset (fig. 1), LOD scores decreased with increasing age, after 51 years.

Heterogeneity Test

The A-test was used to evaluate genetic heterogeneity within the overall group of families (ENVG + VG + late onset), using results for D14S43. The null hypothesis of no linkage was rejected for all three models (table 4). For models 1 and 2, the hypothesis of linkage with heterogeneity (H_2) was significantly favored over the hypothesis of linkage with homogeneity (H_1), while, for model 3, the results for the same comparison were not significant. For model 1, the estimated maximum LOD score (Z_{max}) value was 6.00 ($\hat{\theta}$ = .001), with the fraction of linked families = .20. For model 3, the estimated Z_{max} was 6.88 ($\hat{\theta}$ = .001), with an estimated fraction of linked families = .31. The results are similar to the LOD scores and estimated recombination fraction ($\hat{\theta}$) obtained when only the ENVG families were considered (model 1, Z_{max} = 9.15, $\hat{\theta}$ = .01; model 2, Z_{max} = 5.94, $\hat{\theta}$ = .0; Schellenberg et al. 1992*a*). The probabilities from the A-test for linkage of D14S43 to FAD for each model are given in table 1. No late-onset family had a probability of linkage >.71. When the ENVG and VG families were considered without the late-onset kindreds, the probabilities that a given ENVG kindred is linked to chromosome 14 were greater (table 1).

The M-test was used to test for genetic heterogeneity among the three family groups (ENVG, VG, and late

Table 4

Heterogeneity Analysis

Model and Hypothesis	χ^2	P
1:		
H_2 vs H_1	8.90 (1)	.0014
H_1 vs. H_0	18.74 (1)	<.0001
H_2 vs. H_0	27.64 (2)	<.0001
2:		
H_2 vs. H_1	6.59 (1)	.0051
H_1 vs. H_0	25.10 (1)	<.0001
H_2 vs. H_1	31.69 (2)	<.0001
3:		
H_2 vs. H_1	1.27 (1)	.1299[a]
H_1 vs. H_0	9.53 (1)	.001
H_2 vs. H_0	10.80 (2)	.0023

NOTE.—Results are from the A-test using all VG, ENVG, and late-onset families. Hypotheses tested were as follows: H_2, linkage with genetic heterogeneity; H_1, linkage with genetic homogeneity; and H_0, no linkage (null hypothesis).

[a] Not significant.

onset). Significant results were obtained for all three models (model 1, $\chi^2 = 21.83$, $P = .0001$; model 2, $\chi^2 = 0.00005$; model 3, $\chi_2 = 15.98$, $P = .003$).

APM Analysis

Linkage studies of late-onset AD could potentially be confounded by incorrect specification of the mode of inheritance and/or incorrect parameters for age-at-onset correction. Thus the maximum-likelihood approach used above could potentially give false-negative results. In contrast, the APM method is independent of the mode of inheritance and uses only data from affected individuals. The APM method was applied to test for linkage of FAD to D14S43, D14S53, and D14S52 in the ENVG, VG, and late-onset groups (table 5). For the ENVG families, the null hypothesis of independent segregation of AD with D14S43 and D14S52 was rejected, providing significant evidence in favor of linkage in this group. There was no evidence in support of linkage in the VG families or in the late-onset families when analyzed as one group. However, when the late-onset families were grouped into those with mean age-at-onset range of 60–70 years and those >70 years, evidence for linkage ($P = .03$) to D14S52 was obtained for the group with the relatively earlier onset (table 5). The results in table 5 were obtained using population allele frequencies obtained from Caucasian control subjects (Schellenberg et al. 1992*a*). Since APM analysis is sensitive to perturbations in marker allele frequencies, APM analysis was also performed using marker allele frequencies derived from the entire family group (ENVG + VG + late onset) and frequencies derived from the individual family groups (data not shown). The results obtained were consistent with those presented in table 5.

ASP Analysis

The haplotype sharing in affected siblings was determined in 36 late-onset families; 22 families had two

Table 5

APM Analysis

Family Group[a]	Test Statistics (Significance) for[b] D14S43	D14S52	D14S53
Late-onset:	−2.370 (ns)	.557 (ns)	1.300 (ns)
Mean age at onset 60–70 years	−1.209 (ns)	1.955 (.03)	.021 (ns)
Mean age at onset >70 years	−2.149 (ns)	−1.206 (ns)	1.796 (.05)
VG	.653 (ns)	.705 (ns)	.072 (ns)
ENVG	2.389 (.02)	2.244 (.02)	1.531 (ns)

[a] The following families were analyzed: late-onset families—CSF, MI, P, WLA, CK, RR, LQ, PH, FF, BCR, BEH, EL, FH, QDH, CFD, LJT, SB, KAM, WFL, HTB, FZG, BOB, ORAS, ORCW, OREB, ORDF, ORGK, ORGL, ORHM, ORHW, ORJV, ORKD, ORLA, ORLH, ORLR, ORRH, ORSK, ORMK, ORPK, ORSS, ORWW, HR-XII, XVII, BL HR-IX, and HR-XI; early-onset VG families—HD, KS, R, W, and HB; and ENVG families—L, LH, SNW, V, HR-I, and HR-XV.

[b] Test statistics shown are for the function $f(p) = 1/\sqrt{p}$. The function $f(p) = 1$ also gave significant results for D14S43 in early-onset families and for D14S52 in families with the mean age at onset of 60–70 years. The remainder of the cases, which are shown as nonsignificant for $f(p) = 1/\sqrt{p}$, were also nonsignificant for the other weighing schemes. P values are given in parentheses and were empirically determined through 5,000 iterations. ns = not significant.

Table 6

ASP Analysis of Late-Onset Families

	No. of Haplotypes Shared (IBD)[a]			No. of Sib Pairs Analyzed	No. of Families Analyzed
	2	1	0		
All affected pairs[b]	21	40	16	77	36
Expected[b]	19.25	38.5	19.25	77	
One pair per family[c]	8.49	18.57	9	36	36
Expected[c]	9	18	9	36	

[a] IBD = identity by descent.
[b] $\chi^2 = .766$ (1 df), not significant.
[c] $\chi^2 = .054$ (1 df), not significant.

affected sibs, 11 had three (3 pairs), and 2 had four (6 pairs), and 1 had five (10 pairs). Table 6 shows the overall haplotype-sharing distribution for all pairwise combinations and for one pair per family, based on the average sharing in the sibship. In both cases, the observed distribution is close to the random expectations, thus providing no evidence for linkage in late-onset families. The ENVG, VG, and the two subgroups of late-onset families were not analyzed by ASP because of the small sample size.

Discussion

Maximum-likelihood and nonparametric linkage analysis methods were used to determine whether the chromosome 14 FAD locus is responsible for late-onset FAD. To avoid false-negative results, different genetic models were used in the maximum-likelihood analysis. Model 1 approximates the hypothesis that all late-onset FAD is autosomal dominant and that penetrance is age dependent (Mohs et al. 1987). Model 2 (1% penetrance) assumes that all cases of AD are genetic but differs from model 1 in that a very low penetrance is used in order to reduce dependence of the results on assumptions of the FAD gene carrier status of unaffected individuals. Model 3 assumes that not all AD is genetic. AD is a very common disease in the elderly, with prevalence rates increasing exponentially with age and, in some studies, reaching 47% for individuals ≥ 85 years (Evans et al. 1989). Thus, there may be a substantial number of sporadic cases in the families being studied. Model 3 accounts for this possibility by specifying a cumulative normal age-at-onset distribution for sporadic cases in addition to the genetic cases; the sporadic function was chosen to approximate the observed population prevalence. Since maximum-likelihood analysis is relatively robust to model misspecification and heterogeneity, linkage could also be detected by using these models in the presence of genetic and/or etiologic heterogeneity or a mixed mode of inheritance. However, under all models tested, the maximum-likelihood results were uniformly negative, and strong evidence against linkage of FAD to the region defined by D14S53, D14S43, and D14S52 was obtained ($Z \leq -2.0$) for the late-onset families (tables 2 and 3).

The nonparametric APM method, which is not sensitive to some of the factors which confound linkage analysis, also yielded no evidence for a late-onset chromosome 14 locus when all families were analyzed together. However, the APM-method results for linkage analysis of D14S52 to FAD in the families with mean age at onset between 60 and 70 years was statistically significant. If this result is not due to a chance deviation because of multiple comparisons or possible misspecification of allele frequencies, these analyses suggest that the chromosome 14 FAD locus may be responsible for AD in some of the families with mean age at onset between 60 and 70 years. However, it is unlikely that the chromosome 14 FAD locus is the major gene responsible for AD in the majority of late-onset families studied. An allelic variant of the chromosome 14 locus could make a minor contribution to AD susceptibility, however, if multilocus inheritance is involved in some late-onset FAD.

FAD is clearly genetically heterogeneous. Mutations in the APP gene on chromosome 21 are unambiguously pathogenic for FAD in a subset of early-onset families (Chartier-Harlin et al. 1991; Goate et al. 1991; Murrell et al. 1991; Mullan et al. 1992*a*). A second early-onset locus exists on chromosome 14 (Schellenberg et al.

1992*a*). Recent work confirming the existence of this locus (Mullan et al. 1992*b*; St George-Hyslop et al. 1992; Van Broeckhoven et al. 1992) demonstrates that most early-onset FAD is the result of the chromosome 14 gene. In the study by St George-Hyslop et al. (1992), a number of late-onset families were included. As noted by those authors, most of the evidence for linkage came from early-onset families, and the few late-onset kindreds included had negative linkage results.

Earlier reports of a second early-onset chromosome 21 gene centromeric to the APP gene (St George-Hyslop et al. 1987) appear to have been incorrect, since the families previously reported to be linked to chromosome 21 markers (FAD1, FAD2, FAD3, and FAD4) each show significant positive LOD scores for linkage to chromosome 14 markers (St George-Hyslop et al. 1992). The kindred giving most support for linkage to chromosome 21 was FAD4, which yielded near-significant LOD scores for some chromosome 21 markers (e.g., $Z_{max} = 2.94$, $\hat{\theta} = .15$ for D21S52; $Z_{max} = 0.94$, $\hat{\theta} = .05$ for D21S1/D21S11; and $Z_{max} = 2.19$, $\hat{\theta} = .05$ for D21S13/D21S16) (St George-Hyslop et al. 1990; Tanzi et al. 1991). This same family gave significant positive LOD scores for the chromosome 14 marker D14S55 ($Z_{max} = 5.21$, $\hat{\theta} = .05$) and suggestive positive values for D14S53 ($Z_{max} = 2.52$, $\hat{\theta} = 0$) and D14S43 ($Z_{max} = 2.17$, $\hat{\theta} = 0$) (St George-Hyslop et al. 1992). Since the VG families do not show evidence for linkage to chromosome 14, an additional early-onset FAD locus may exist. The evidence presented here and elsewhere (Pericak-Vance et al. 1988; Schellenberg et al. 1991; Kamino et al. 1992) demonstrates that neither the APP gene nor the chromosome 14 locus is a major contributor to late-onset FAD.

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