Letters to the Editor

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A Locus for the Nystagmus-Associated Form of Episodic Ataxia Maps to an 11-cM Region on Chromosome 19p

To the Editor:

Episodic ataxia (EA) is a rare neurological disorder characterized by attacks of generalized ataxia and near-normal neurological function between attacks. Most inherited cases are the result of an autosomal dominant condition with unknown neuropathology. It is heterogeneous and includes at least two distinct forms. In EA-1, attacks last minutes and interictal myokymia may be present. In EA-2, attacks may last hours and interictal nystagmus may occur.

We reported linkage in four EA-1 families to chromosome 12p13 (Litt et al. 1994) and identified mutations in these families in a K^+ channel gene, *KCNA1* (Browne et al. 1994). Recently, we reported linkage in two EA-2 families to a 30-cM region on chromosome 19p (Kramer et al. 1994*a*, 1994*b*). While developing collaborations for additional EA family material, we found that investigators at the University of California, Los Angeles, were studying members of the same two families and one additional kindred. This report is based on these three families.

Pedigrees are shown in figure 1. Detailed clinical descriptions are given in Zasorin et al. (1983), Gancher and Nutt (1986), and Baloh and Winder (1991). We identified individuals as affected if they reported a history of intermittent attacks of incoordination, staggering, or slurred speech and/or if they exhibited neurological signs suggesting cerebellar dysfunction. Other features supportive of a diagnosis of EA-2 include nystagmus or trunkal instability, beneficial response to acetazolamide, and family history.

After obtaining informed consent of family members, we isolated DNA from peripheral blood (Bell et al. 1981) and typed four microsatellite markers (Litt et al. 1993) with the following map locations (Gyapay et al. 1994): D19S413–5 cM–D19S221–6 cM–D19S226/D19S415.

We used the MLINK option in FASTLINK (Schaffer et al. 1994) for two-point linkage analysis, and assumed autosomal dominant inheritance of a rare gene (frequency .0001). Family 3 shows evidence of incomplete penetrance, since unaffected individual III-2 has an affected father and an affected child. We estimated penetrance to be .95. Since onset of EA occurs by late adolescence and all unaffected individuals for whom we have data were older than 15 years, no correction for age-dependent penetrance was required.

We identified 23 individuals affected with EA (see table 1). Most affected members described recurrent attacks of limb and trunkal incoordination. The frequency and severity of attacks varied substantially, to the extent that treatment with acetazolamide was not warranted in 11 cases. Two members of family 1 (I-2 and II-2) and one of family 2 (II-6) denied attacks of ataxia but were identified as affected on the basis of the presence of nystagmus and/or trunkal ataxia. In families 1 and 3, nystagmus was a typical feature in all but two individuals, III-10 and III-9 in family 1; in family 2, only two affected individuals, I-1 and II-4, exhibited nystagmus. Acetazolamide was effective in reducing the frequency of attacks in nearly all cases in which it was tried; the exception was III-3 in family 1, who experiences frequent and severe attacks of ataxia and no relief with acetazolamide.

The strongest evidence for linkage occurred at D19S221 (total lod score = 5.07 at $\theta = .01$), which was the only locus with no obligate crossovers in any of the three kindreds. We also obtained significant evidence for linkage to both D19S413 and D19S226/D19S415, with total lod scores > 3.00 at $\theta = .10$.

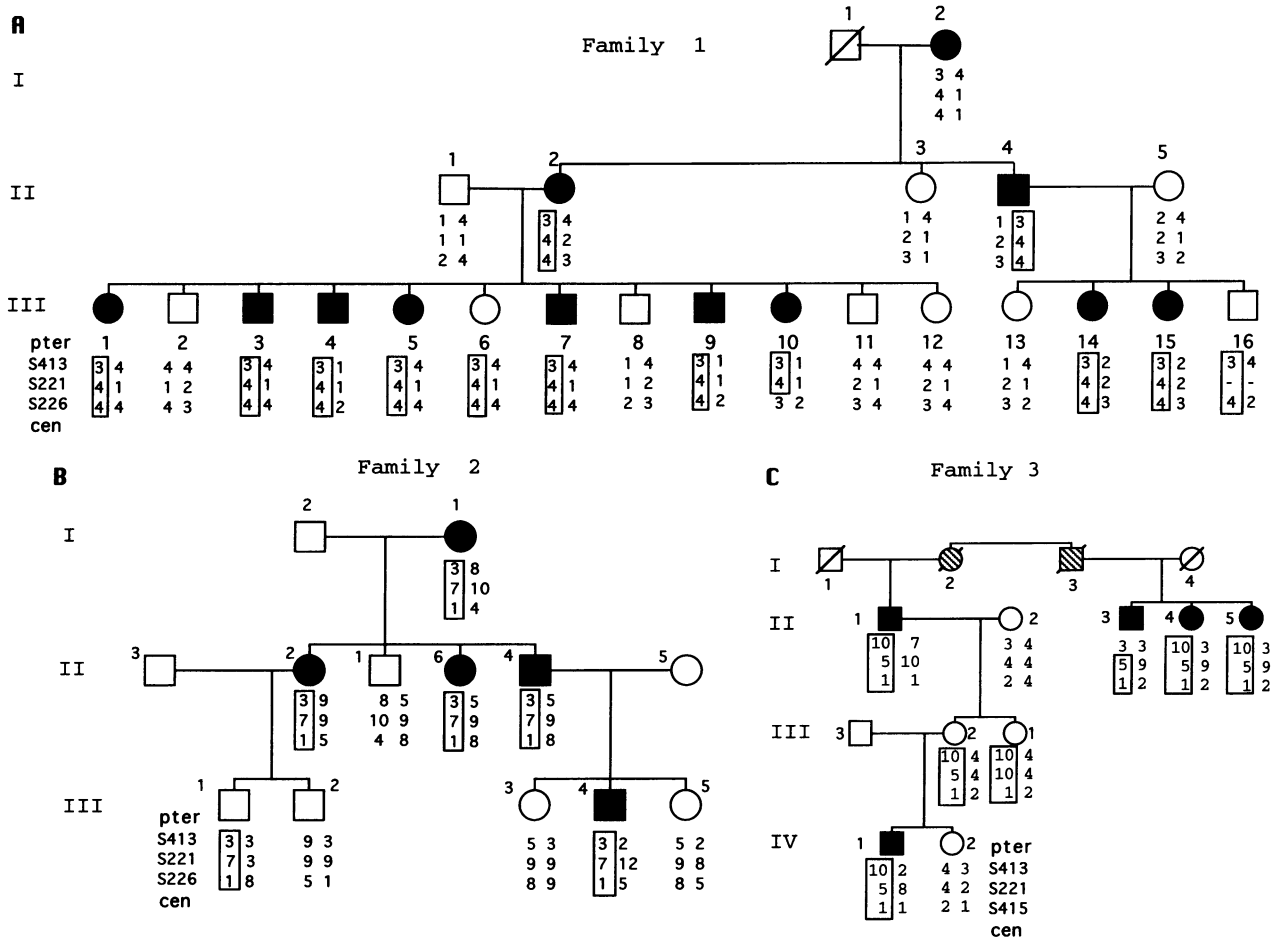

Figure 1 Pedigrees of families studied and haplotypes of chromosome 19p loci. Blackened symbols denote affected individuals who have been examined; hatched symbols denote individuals affected by history but not examined; clear symbols denote unaffected individuals. Genotypes are listed in the order given by the maps to the left of each pedigree. Samples were available for all individuals for whom a haplotype is indicated. The haplotype of the disease-bearing chromosome is boxed.

Haplotype data are given in figure 1. Critical crossovers occurred on the disease-bearing chromosomes of two affected individuals. III-10 (family 1) shows a D19S221-D19S226 crossover; II-3 (family 3) shows a D19S413-D19S221 crossover. Affected status was reconfirmed in both cases. These data indicate that a gene for EA-2 is located within an 11-cM region flanked by D19S413 and D19S226.

Four individuals carry a disease-bearing chromosome but show no symptoms. In family 1, III-6 and III-16 were examined at 27 years and 16 years, respectively; in family 3, III-2 was examined within the last 6 months; in family 2, III-1 was interviewed recently by telephone. None show any signs or symptoms of EA. This suggests that the penetrance estimate we used was probably too high and that environmental factors (or other modifying genes) play some role in eventual manifestation of the disorder.

von Brederlow et al. (1995) reported linkage in two EA-2 families to a 12-cM region on 19p between D19S391 and D19S179 (Cooperative Human Linkage Center map; Buetow et al. 1994). This overlaps nearly the entire region we report here. Vahedi et al. (1995) reported linkage in a large EA-2 family to a 30-cM region on 19p flanked by D19S216 and D19S215; this includes the entire 11-cM region reported here.

Joutel et al. (1993) mapped a gene for familial hemiplegic migraine (FHM) to the region around D19S221 in two large French families. Several individuals had interictal nystagmus, two had cerebellar atrophy, and one had cerebellar ataxia; none had EA. Although none of the EA-2 individuals we report had hemiplegic migraine, some reported headaches during ataxic episodes. It may be that EA-2 and some FHM represent different mutations in the same locus on 19p.

All of the autosomal dominant periodic neurological diseases whose causative genes have been characterized are associated with ion channel defects (Ashcroft and

Table 1

Clinical Data on 23 Affected Individuals with Episodic Ataxia

Individual	Age at Last Exam (years)	Sex	Age at Onset of Attacks	Nystagmus	Acetazolamide-responsive	Headache
Family 1:						
I-2	88	F	...[a]	Yes	...[b]	No
II-2	59	F	...[a]	Yes	...[b]	No
II-4	54	M	17	Yes	Yes	Yes
III-1	36	F	Adolescence	Yes	...[b]	Yes
III-3	38	M	Adolescence	Yes	No	No
III-4	30	M	13–15	Yes	...[b]	?
III-5	29	F	12	Yes	Yes	No
III-7	26	M	12	Yes	...[b]	No
III-9	24	M	Adolescence	No	...[b]	?
III-10	18	F	Adolescence	No	Yes	Yes
III-14	24	F	14	Yes	...[b]	Yes
III-15	23	F	16	Yes	...[b]	No
Family 2:						
I-1	85	F	Childhood	Yes	...[b]	No
II-2		F	Adolescence	No	Yes	Yes
II-4	68	M	15	Yes	Yes	Yes
II-6	56	F	...[a]	No	...[b]	No
III-4	33	M	12	No	Yes	No
Family 3:						
II-1	58	M	3	Yes	Yes	Yes
II-3	69	M	29	Yes	Yes	No
II-4	55	F	13	Yes	...[b]	No
II-5	52	F	10	Yes	Yes	No
IV-1	7	M	2	Yes	Yes	Yes

[a] No history of ataxic episodes; affected status based on nystagmus and trunkal ataxia on examination in I-2 and II-2 and on history of attacks of vertigo in II-6.

[b] Frequency and/or severity of attacks did not warrant administration of acetazolamide.

Roper 1993). Furthermore, patients with mutations in genes encoding cation channels frequently respond to acetazolamide and other carbonic anhydrase inhibitors (Ptacek 1994). EA-2 shares this characteristic. In keeping with the pharmacological behavior of the drug, two families with acetazolamide-responsive EA-2 show abnormally high intracerebellar pH, which was normalized by the drug treatment (Bain et al. 1992). Because they are the largest and most diverse class of ion channels known and because of the precedent furnished by our discovery of abnormal K^+ channels in EA-1, K^+ channels are candidate genes for EA-2. Genes coding for sodium or calcium channels are also of interest. The current version of the Genome Database does not show any ion channel genes localized to human chromosome 19p. However, it seems likely that many ion channel genes remain to be mapped; any that localize to 19p13 will be candidate genes for EA-2.

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The Spinocerebellar Ataxia 2 Locus Is Located within a 3-cM Interval on Chromosome 12q23-24.1

To the Editor:

The autosomal dominant cerebellar ataxias (ADCA) are a clinically heterogeneous group of neurodegenerative disorders characterized by a predominantly cerebellar syndrome of onset with gait ataxia, dysarthria, dysmetria, and dysdiadochokinesia. Pathologically, the disorders are characterized by premature neuronal loss in the cerebellar cortex and the inferior olivary and pontine nuclei, with degeneration of the spinal cord.

Genetic heterogeneity has been established, with disease loci assigned to chromosomes 6q (Jackson et al. 1977), 11 (Ranum et al. 1994), 12q (Twells et al. 1993), 14q (Takiyama et al. 1993; Stevanin et al. 1994) and 16q (Gardner et al. 1994). Two of these genes have been isolated, and the mutation mechanism has been shown to be unstable $(CAG)_n$ motifs present within coding sequence (Orr et al. 1993; Kawaguchi et al. 1994).

Elsewhere, we have assigned the spinocerebellar ataxia 2 locus to chromosome 12q23-24.1, within a 31-cM interval flanked by the loci D12S58 and PLA2 (Twells et al. 1993). Although initially ascertained in a potential founder population from the Holguin province, Cuba, mutation at this locus is not unique to the Cuban kindred. Linkage to SCA2 has been demonstrated in pedigrees from Europe, Japan (Sasaki et al. 1993), and North America (Lopes-Cendes et al. 1994), the latter study serving to refine the candidate region to a 16-cM interval.

We report here genetic analysis undertaken between SCA2 and nine microsatellite loci known to span 8 cM within this interval. A total of 176 individuals, including 121 affected members from 16 pedigrees, were included in the analysis. A description of the phenotype has been reported elsewhere (Orozco-Diaz et al. 1990). The order and sex-averaged distance (in cM) between these markers is as follows: cen–D12S353–(0.00)–D12S330–(0.02)–D12S84–(0.00)–D12S105–(0.00)–AFM240we1–(0.03)–AFM128yf1–(0.00)–AFM312yb1–(0.01)–D12S354–(0.02)–D12S79–qter. Primer sequences were obtained from the Genome Database or by one of us (J.W.). Microsatellite analysis was performed following PCR amplification incorporating 100 ng genomic DNA, 50 pmol each of the forward and reverse primer, 200 μM dGTP, dCTP, dTTP, 25 μM dATP, 10 μCi ^{35}S-dATP and 0.25 U *Taq* polymerase (Dynazyme) in a standard 25 μl reaction. Amplification was carried out following an initial denaturation at 95°C for 7 min