X linked spastic paraplegia (SPG2): clinical heterogeneity at a single gene locus

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Abstract

X linked hereditary spastic paraplegia is a rare condition that has been divided into two forms (the pure spastic form and the complicated form) as a function of clinical course and severity. A gene for pure hereditary spastic paraplegia (SPG2) has been mapped to the proximal long arm of the X chromosome (Xq21) by linkage to the *DXS17* locus, while a gene for a complicated form of the disease has been mapped to the distal long arm by linkage to the *DXS52* locus (Xq28).

Here we report on the mapping of a gene for complicated hereditary spastic paraplegia to the Xq21 region by linkage to the probe S9 at the *DXS17* locus ($Z = 5$ at $\theta = 0.04$) in a three generation pedigree. Multipoint linkage analysis supports the distal location of the disease gene with respect to the *DXYS1-DXS17* block (cen-*DXYS1-DXS3-DXS17-SPG2*-tel).

The observation of a complicated form of spastic paraplegia mapping to Xq21 raises the difficult issue of variable phenotypic expression, allelic heterogeneity, or even close proximity of two genes for hereditary spastic paraplegia in this region. However, since our study provides clinical evidence for intrafamilial heterogeneity in complicated X linked spastic paraplegia, the present data support the hypothesis of variable clinical expression of a single gene at the *SPG2* locus, as previously suggested for *SPG1*. Finally, we report here what we believe to be the first evidence of clinical expression in heterozygous carriers, a feature that is relevant to genetic counselling in at risk females.

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X linked hereditary spastic paraplegia (HSP) is a rare condition whose clinical heterogeneity has long been recognised.¹ While pure spasticity of the lower limbs has been reported in some pedigrees,^{2,3} most patients reported were found to display a more complicated neurological profile including cerebellar ataxia, sensory loss, nystagmus, optic atrophy, congenital malformations, and mental retardation.⁴⁻¹⁰

However, two distinct loci for X linked hereditary spastic paraplegia, *SPG1* and *SPG2*, have been identified by linkage analysis in informative families. *SPG1* maps to the distal long arm of the X chromosome (Xq28),⁸ while *SPG2* maps more proximally (Xq21).³

Since *SPG1* was mapped in a large pedigree with complicated spastic paraplegia and *SPG2*

in a family with pure spastic paraplegia, it was thought that *SPG1* might account for the complicated form and *SPG2* for the pure spastic form of the disease. Subsequently, however, a gene for complicated spastic paraplegia was also mapped to the Xq21 region suggesting either allelic heterogeneity at the *SPG2* locus or close proximity of two loci for spastic paraplegia in this region (one pure, one complicated).⁹

Here we report on a large pedigree with complicated hereditary spastic paraplegia whose disease gene maps to the proximal long arm of the X chromosome (Xq21) and we provide additional evidence for intrafamilial heterogeneity of this condition. Moreover, we present what we believe to be the first evidence of clinical expression in heterozygous carriers.

Patients and methods

PATIENTS

The present study was carried out in a three generation pedigree (fig 1) including 11 affected males and six obligate carriers. Extensive clinical examination was possible in 35 subjects (10 affected males, four obligate carriers, 21 relatives). Additional medical records and information were obtained in one affected male and two obligate carriers.

The clinical findings in three affected males and two expressing females (including an obligate carrier) are presented below. Clinical data on other affected males and heterozygous carriers are shown in tables 1 and 2.

CASE 1

The proband (IV.3), born in 1975, was first referred for abnormal gait at 17 months of age. Clinical evaluation showed mild spasticity of the lower limbs associated with severe ataxia and multidirectional nystagmus. Generalised seizures occurred at the age of 10 years, spastic paraplegia worsened thereafter, and he became wheelchair bound at the age of 13 years. At 14 years of age, he had spastic paraplegia and cerebellar syndrome of the upper extremities with dysarthria and nystagmus. His intelligence was impaired (IQ of 41 at the age of 9 years) and special schooling was required from the age of 5 years.

CASE 2

Patient IV.5, the brother of the proband, was born in 1981. Nystagmus and hyperreflexia were first noted at the age of 17 months. At 30 months, he had pyramidal signs of the lower

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Figure 1 Pedigree of family.

Table 1 Clinical features in affected males.

Cases	Age at study (y)	First symptoms	Neurological signs
II.3	NE	Nystagmus and abnormal gait in early childhood	Spastic gait at 20 y; required crutches. Sudden death at 34 y
III.3	40	Started walking at 3 y. Nystagmus noted early	Spastic and ataxic gait at 6 y; wheelchair bound at 20 y. Pyramidal signs in lower limbs. Normal upper limbs. Horizontal nystagmus. Normal intelligence
III.7	26	Started walking at 3 y	Spastic and ataxic gait. Wheelchair bound at 16 y. Pyramidal signs in lower limbs. Normal upper limbs. Mild horizontal nystagmus. Normal intelligence
III.12	33	Abnormal gait, pes cavus, nystagmus, and spasticity noted at 5 y	Requires crutches. Pyramidal signs in lower limbs. Mild tremor in upper limbs. Horizontal nystagmus. Normal intelligence
III.13	34	Started walking at 20 mth. Abnormal gait, nystagmus, spasticity, and cerebellar signs noted at 4 y	Wheelchair bound. Pyramidal signs in lower limbs. Mild cerebellar signs in upper limbs. Normal intelligence
III.19	31	Nystagmus noted at 6 mth. Abnormal gait from early infancy	Wheelchair bound. Pyramidal signs in lower limbs. Mild tremor in upper limbs. Dysarthria. Mild mental impairment
III.22	30	Nystagmus and abnormal gait noted at 3 y	Abnormal gait. Pyramidal signs in lower limbs. Horizontal nystagmus. Normal intelligence
III.25	13	Total absence of abnormal signs	Detailed in text
IV.3	14		Detailed in text
IV.5	8		Detailed in text
IV.18	1	Born at 30 weeks' gestation, birth weight 1800 g. Nystagmus first noted at 11 mth	At 1 y hypotonia, left Babinski's sign, horizontal nystagmus

NE = not examined.

Table 2 Clinical features in heterozygous females.

Case	Status	Age at study (y)	Abnormal gait	Spastic paraparesis	Pes cavus	Comments
I.4	OC	NE	+++			Required wheelchair at 50 y
II.2	OC	46	+++	++	+	Abnormal gait from 20 y
II.5	OC	55	—	—	—	
II.6	OC	53	—	—	—	Brisk reflexes in lower limbs
III.4	R	40	—	—	—	
III.5	OC	28	—	+	+	
III.9	R	20	++	++	+	Borderline intelligence
III.14	OC	24	—	+	+	
III.15	R	23	—	—	—	
III.24	R	23	—	—	—	

NE = not examined, OC = obligate carrier, R = female considered at risk of carrying the disease on the basis of her status at DXS17.

limbs with marked ataxia, mild dysmetria, tremor of the upper extremities, and multidirectional nystagmus. Heel contracture required heel cord lengthening at 5 years. At 8 years he had severe spastic paraplegia with cerebellar syndrome of the lower limbs and persistent cerebellar signs of the upper limbs. His visual acuity was markedly reduced (20/100) but no optic atrophy was noted. His intelligence and schooling were normal.

CASE 3

Patient III.25, born in 1976, did well during the first 12 years of life. His intelligence and schooling were normal; his gait was also normal and he played sport regularly. Extensive neurological evaluation at 13 years of age, however, showed evidence of mild involvement of the pyramidal tract with very brisk reflexes of the lower limbs without Babinski's sign.

CASE 4

Patient III.5, born in 1950, is an obligate carrier as she is the mother of the proband (IV.3) and the sister of two affected males (III.3 and III.7). At 38 years of age, her gait and intelligence were normal but she had pes cavus and extensive neurological evaluation showed evidence of brisk deep tendon reflexes of the lower limbs with bilateral Babinski's sign.

CASE 5

Patient III.9 is the maternal aunt of the proband, born in 1968. She did well during the first years of life and her intelligence and coordination were normal. However, she had an abnormal gait with pes cavus, spastic para-

Table 3 Pairwise lod scores.

Locus	Probe	Recombination fraction										
		0.0	0.01	0.02	0.03	0.04	0.05	0.06	0.07	0.08	0.1	0.2
<i>DXS17</i>	S9	−∞	4.71	4.91	4.98	5.00	4.99	4.96	4.91	4.86	4.73	3.85
<i>DXYS1</i>	PDP34	−∞	0.64	1.17	1.45	1.63	1.76	1.84	1.90	1.95	1.99	1.82
<i>DXS3</i>	P19.2	−∞	1.37	1.89	2.15	2.32	2.42	2.49	2.54	2.57	2.58	2.26
<i>DXS52</i>	St14	−∞					−4.68				−2.61	−0.91

paresis, and leg muscle wasting at 22 years of age.

Methods

Total DNA was extracted from circulating leucocytes, digested using restriction enzymes, and submitted to gel electrophoresis for Southern blotting. Membranes were hybridised using ^{32}P -dCTP labelled DNA probes at the polymorphic loci *DXYS1* (probe pDP34), *DXS3* (probe p19.2), *DXS287* (probe pYNH3), *DXS17* (probe S9), and *DXS52* (probe St14).¹¹

Linkage analysis was performed on a personal computer using the LINKAGE program version 4.9. Pairwise lod scores were calculated using the MLINK option.¹² Multipoint linkage analysis was performed using the LINKMAP program.¹³ Genetic distances between markers were obtained from a previous study.¹⁴

Results

A total of 35 subjects including 10 affected males, seven healthy males born to heterozygous mothers, five obligate carriers, and 13 relatives were tested. Table 3 shows the pairwise lod scores between the disease locus and loci *DXYS1*, *DXS3*, *DXS17*, and *DXS52*. The maximum lod score value ($Z=5$ at $\theta=0.04$) was obtained with probe S9 at locus *DXS17* (Xq21.3–q22).

Positive lod score values were also obtained with two other polymorphic probes for the q21 region, namely P19.2 and pDP34 at loci *DXS3* and *DXYS1* respectively (table 3). Probe pYNH3 at locus *DXS287* was not informative. Two recombination events were observed in subjects III.9 and III.13 (fig 1).

Figure 2 Multipoint linkage analysis in SPG2 disease. Likelihood estimates are given in log base 10. Maximum location score of 5.22 was obtained between *DXS17* and the telomere at 3.4 cM from *DXS17*.

In subject III.9 (a female expressing the disease) recombination occurred between *DXYS1* and *DXS3–DXS17*. In subject III.13 (an affected male) recombination occurred between *DXS3* and *DXS17*.

Negative lod score values were obtained with probe St14 excluding the disease from close proximity to the *DXS52* locus ($Z=-2$ at $\theta=0.12$).

The location score method was used to estimate the position of the disease locus. The maximum likelihood estimate for location of the SPG2 gene was between *DXS17* and the telomere (maximum location score of 5.22 at 3.4 cM from *DXS17*) (fig 2). Finally, multipoint linkage analysis allowed the placing of the disease locus with respect to polymorphic loci: cen-*DXYS1–DXS3–DXS17–SPG2*-tel.

Discussion

The present study reports on the mapping of a gene for a complicated form of X linked hereditary spastic paraplegia to the proximal long arm of the X chromosome (Xq21) by close linkage to the *DXS17* locus in a three generation pedigree. This report is of interest for several reasons. First, it presents what we believe to be the first clinical evidence for mild expression of the disease in heterozygous carriers and this observation is relevant when considering the genetic status of at risk females in affected pedigrees.

Second, this study gives additional support to the variable expression of the disease gene among affected subjects. Indeed, a broad spectrum of clinical phenotypes was observed in affected males, ranging from severe spasticity with nystagmus, cerebellar syndrome, and mental retardation to mild pyramidal tract involvement with normal gait and intelligence. This intrafamilial heterogeneity has been previously noted by Johnston and McKusick in a large pedigree of 17 affected males in whom spastic paraplegia was associated, to various degrees, with sensory loss, nystagmus, optic atrophy, dysarthria, and mental retardation.⁵ Similarly, Goldblatt *et al*⁹ reported variable phenotypes of X linked HSP in a large kindred in which pyramidal signs were associated with nystagmus, optic atrophy, mild ataxia of the arms, and intellectual impairment. Our family manifests a complicated X linked HSP very similar to that described by Johnston and McKusick⁵ and Goldblatt *et al*⁹ and it appears therefore that clinical heterogeneity is a consistent feature in this form of X linked spastic paraplegia.

Finally, the present study establishes linkage of the disease gene to the *DXS17* locus ($Z=5$ at $\theta=0.04$) in one single family and gives additional support to the presence of a

gene for complicated HSP on the proximal long arm of the X chromosome (Xq21). Since uncomplicated forms of the disease (that is, isolated spasticity of the lower limbs) have been previously shown to be linked to the same locus ($Z = 4.48$ and $Z = 4$ at $\theta = 0$ for *DXS287* and *DXS17* respectively),³ our results along with those of Goldblatt *et al*⁹ raise the difficult issue of either allelism at the *SPG2* locus or close proximity of two disease genes, one for pure hereditary spastic paraplegia (*SPG2*) and another one for a complicated form of HSP.

In connection with this, it is worth remembering that remarkable heterogeneity has also been reported at the *SPG1* locus. For this reason, some authors^{15,16} tend to favour the view that different mutations at the same locus account for both the complicated spastic paraplegia and MASA syndrome phenotypes (mental retardation, aphasia, shuffling gait, and adducted thumbs)¹⁷ whose gene has been mapped to the same Xq28 region.^{18,19}

More generally, it is tempting to hypothesise that different mutations of a single gene could account for the remarkable clinical heterogeneity of X linked hereditary spastic paraplegias, as recently shown for autosomal and X linked genetic diseases.²⁰

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