LETTER TO JMG

Deletion of the SIM1 gene (6q16.2) in a patient with a Prader-Willi-like phenotype

L Faivre, V Cormier-Daire, J M Lapierre, L Colleaux, S Jacquemont, D Geneviève, P Saunier, A Munnich, C Turleau, S Romana, M Prieur, M C De Blois, M Vekemans

A part from Prader-Willi syndrome, which is a well delineated imprinting disorder of the 15q11-q12 region, other chromosome anomalies have been described in a small number of patients with features reminiscent of Prader-Willi syndrome, including hypotonia, progressive obesity, small extremities, and delayed developmental milestones. Among these chromosome anomalies are some cases of interstitial deletion of chromosome 6q16.2 and haploinsufficiency of the SIM1 gene (6q16.2) has been proposed as a candidate gene for obesity. Here, we report a fifth case of Prader-Willi-like phenotype associated with an interstitial chromosome 6q deletion (6q16.1-q21) detected only by high resolution banding techniques. This suggests that a subgroup of patients with features reminiscent of Prader-Willi syndrome and an interstitial deletion of chromosome 6q16.2 could be delineated.

CASE REPORT

The proband was the only child of a 27 year old mother and a 32 year old father. Intrauterine growth retardation, oligohydramnios, and a left club foot were noted during the third trimester of pregnancy. He was born at term after a normal delivery. His growth parameters were weight 2350 g (−2.5 SD), length 47 cm (−1.5 SD), and OFC 33 cm (−1.5 SD). He was described as floppy and had feeding difficulties in early infancy. He sat at the age of 2 years, walked at 3 1/2 years, and had no speech when we first saw him aged 5 years. Excessive weight gain began at 3 years, with a big appetite and food seeking behaviour. There were no sleep disturbances. His behaviour was hyperactive, with a short attention span and intolerance to frustration. Interaction with other children was non-existent.

At physical examination, weight was +5.5 SD, height was −1 SD, and OFC was +2.5 SD. He had generalised obesity, slightly dysmorphic features including a square and flat face, a large forehead with a protruding metopic ridge, small palpebral fissures, mild strabismus, a thin nose, and thin lips (fig 1). The ears were low set with very small lobes. The hands and feet were small with low implanted thumbs, the external genitalia were normal, and a dry skin with livedo was noted. No malformations were found. Clumsiness and a wide based gait were noted at neurological examination. Metabolic screening and fragile X studies were normal, as was a cerebral MRI. Bone age was 3 1/2 years at the age of 5. Psychometric evaluation showed performance at a 2 1/2 year level (DQ = 50).

Cytogenetic and molecular analyses

In order to rule out Prader-Willi syndrome, a methylation assay of the SNRPN gene and a FISH study with the SNRPN probe were performed. Both analyses showed normal results, as well as standard cytogenetic and telomere analyses using microsatellite polymorphic markers. However, high resolution banding analysis performed on cultured peripheral blood lymphocytes using R and G banding techniques showed a 46,XY, del(6)(q16.1q21) karyotype (fig 2A). Karyotypes of the parents were normal. Comparative genomic hybridisation study confirmed the deletion (fig 2B). Molecular studies using microsatellite polymorphic markers D6S1709, D6S1580, and D6S447 (6q16.3-q21) were fully informative and showed that the 6q deletion was paternal in origin. In addition, molecular studies using an intragenic microsatellite polymorphic marker within the SIM1 gene (primers 5′-GGCTGGGCTCAACTCGG-3′ and reverse 5′-GATCCACGAGCAAGAG-3′) were fully informative and showed that the SIM1 gene was deleted (fig 2C).

Figure 1  Picture of the child at the age of 5 years. Note generalised obesity, slightly dysmorphic features including a square and flat face, large forehead with protruding metopic ridge, small palpebral fissures, mild strabismus, thin nose, low set ears and thin lips, and small hands and feet.

subject. Indeed, it has been shown in the mouse that the hypothalamus, could be responsible for obesity in their
the melanocortin 4 receptor in the paraventricular nuclei of deficiency of
SIM1
appear to affect any transcription unit on 1p22.1, it disrupted
translocation breakpoints. While the translocation did not
are summarised in table 1.
found in 2/5 and 3/5 patients, respectively. These differences
seizures, hearing loss, Arnold-Chiari malformation) were
as neurological abnormalities (polygyria, leucomalacia,
(bicuspid aortic valve, aortic stenosis, right branch block) as
excessive appetite was seen in only two patients and cardiac
However, there are also distinct differences. For example,
ity, hypotonia, short extremities, and developmental delay.
A Prader-Willi-like phenotype showed that they all shared the
following features with Prader-Willi syndrome patients: obes-
some 1p22.1 and 6q16.2 has been reported in a patient with
(figure 3). This suggests that the 6q16.2 subband as well as patients with distal deletion of
obesity related phenotypes on distal chromosome 6q
is also deleted in our patient. Therefore, this observation
heritage suggest that in patients with 6q duplication and obesity
In conclusion, this observation suggests that in patients
with a Prader-Willi phenotype and a deletion distal to the
imprinting and obesity in chromosome 6q deletion remains
Further molecular studies of similar cases might help to resolve the possible causal relationship between
imprinting and obesity.
In conclusion, this observation suggests that in patients
with a Prader-Willi phenotype and a normal cytogenetic/
molecular study of the 15q11-q12 region, deletion of the
6q16.2 region and the SIM1 gene in particular should be
looked for.
ACKNOWLEDGEMENTS
We thank M Pick and C Ozilou for their excellent technical work and
Y Deris for his photographic assistance.

DISCUSSION
Here we report on the fifth case of chromosome 6q deletion in
association with a Prader-Willi-like phenotype. A review of
five patients with an interstitial chromosome 6q deletion and
a Prader-Willi-like phenotype showed that they all shared the
following features with Prader-Willi syndrome patients: obesity, hypotonia, short extremities, and developmental delay. However, there are also distinct differences. For example, excessive appetite was seen in only two patients and cardiac (bicuspid aortic valve, aortic stenosis, right branch block) as well as neurological abnormalities (polygyria, leucomalacia, seizures, hearing loss, Arnold-Chiari malformation) were found in 2/5 and 3/5 patients, respectively. These differences are summarised in table 1.
Interestingly, four out of five patients described have a 6q16.2 deletion (figure 3). This suggests that the 6q16.2 subband could be regarded as a region of interest for obesity related genes. In addition, a balanced translocation between chromosomes 1p22.1 and 6q16.2 has been reported in a patient with profound obesity. The authors cloned and sequenced both translocation breakpoints. While the translocation did not appear to affect any transcription unit on 1p22.1, it disrupted the SIM1 gene on 6q16.2. They hypothesised that haploinsufficiency of SIM1, possibly acting upstream or downstream of the melanocortin 4 receptor in the paraventricular nuclei of the hypothalamus, could be responsible for obesity in their subject. Indeed, it has been shown in the mouse that the Sim1 gene is expressed in the central nervous system where it has
an important role in the development of the supraoptic and the paraventricular nuclei of the hypothalamus. In addition, there is good evidence from anatomical and pharmacological studies that these nuclei are involved in the regulation of body weight as these neurones express the melanocortin-4 receptor and appear to be a target of alpha melanocyte stimulating hormone, which inhibits food intake. Interestingly, the SIM1 gene is also deleted in our patient. Therefore, this observation provides further support for the hypothesis that haploinsufficiency of the SIM1 gene might be responsible for the obesity observed in our patient.
There is also some evidence in published reports that other chromosome 6q loci might carry obesity related genes, namely: (1) a patient reported by Stein et al with a Prader-Willi-like phenotype and a deletion distal to the subband 6q16.2 as well as patients with distal deletion of chromosome 6q and obesity alone; (2) the observation of patients with 6q duplication and obesity; and (3) the discovery of a major locus for fasting insulin concentrations and insulin resistance with strong pleiotropic effects on obesity related phenotypes on distal chromosome 6q (figure 3).
Finally, there is good evidence that some chromosome 6q genes are submitted to genomic imprinting (http://www.geneimprint.com). For example, transient neonatal diabetes mellitus is associated with paternal uniparental disomy for chromosome 6 or paternal duplication of chromosome 6q24. In our report, the deletion was of paternal origin, as was the duplication of chromosome 6q24.3-q27 in the patient reported by Smith et al. However, the relationship between imprinting and obesity in chromosome 6q deletion remains unknown. Further molecular studies of similar cases might help to resolve the possible causal relationship between imprinting and obesity.

Table 1  Clinical features observed in Prader-Willi syndrome and five cases of interstitial deletion of chromosome 6q associated with Prader-Willi-like phenotype

<table>
<thead>
<tr>
<th>Feature</th>
<th>Patients with interstitial 6q deletion</th>
<th>Patients with Prader-Willi-like phenotype</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hypotonia</td>
<td>+</td>
<td>4/4</td>
</tr>
<tr>
<td>Feeding problems in infancy</td>
<td>+</td>
<td>3/3</td>
</tr>
<tr>
<td>Obesity</td>
<td>+</td>
<td>5/5</td>
</tr>
<tr>
<td>Facial dysmorphism</td>
<td>Mild</td>
<td>Moderate</td>
</tr>
<tr>
<td>Hypogonadism</td>
<td>+</td>
<td>4/5</td>
</tr>
<tr>
<td>Developmental delay</td>
<td>Mild</td>
<td>Severe</td>
</tr>
<tr>
<td>Hyperphagia</td>
<td>+</td>
<td>2/5</td>
</tr>
<tr>
<td>Short extremities</td>
<td>+</td>
<td>5/5</td>
</tr>
<tr>
<td>Cardiac defects</td>
<td>-</td>
<td>2/5</td>
</tr>
<tr>
<td>CNS abnormalities</td>
<td>-</td>
<td>3/5</td>
</tr>
</tbody>
</table>

Figure 2  (A) High resolution banding of chromosome 6, showing the non-deleted chromosome on the left and the deleted chromosome on the right. (B) Comparative genomic hybridisation studies confirming the 6q deletion. (C) Molecular analysis using intragenic polymorphic microsatellite marker of the SIM1 gene showing the deletion of paternal origin (F: father, M: mother, C: child).
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REFERENCES


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Motor neurone disease includes a heterogeneous group of disorders with motor neurone involvement, such as amyotrophic lateral sclerosis, progressive muscular atrophy, progressive bulbar palsy, and primary lateral sclerosis. Amyotrophic lateral sclerosis is the most common adult onset form of motor neurone disease and involves the lower and upper motor neurones. It is characterised by progressive muscle weakness and atrophy, with fasciculations associated with hyperreflexia and spasticity. One of the proposed mechanisms for amyotrophic lateral sclerosis is degeneration of the motor neurone because of abnormal levels of toxic products that accumulate in the cell. Death usually occurs by respiratory failure about 2–3 years after the first symptoms. About 10% of cases are familial amyotrophic lateral sclerosis, and several loci have been associated with this disease. To date, the only two genes identified have been the zinc–copper superoxide dismutase 1 (SOD1) gene, which is located on chromosome 21 (ALS1, MIM105400), and the Alsin gene, which is located at 2q33 (ALS2, MIM 205100). Autosomal dominant forms of amyotrophic lateral sclerosis also have been linked to 18q21 (ALS3, MIM 606640), 9q34 (ALS4, MIM 602433), and 15q15.1–q21.1 (ALS5, MIM 602099) and amyotrophic lateral sclerosis–frontotemporal dementia (MIM 105550) to 9q21-22. Moreover, mutations in Dynein are associated with motor neurone degeneration and defects in retrograde transport. This gene acts in the cellular division, trafficking, and transport of several proteins, such as SOD1.

More recently, two new loci have been associated with amyotrophic lateral sclerosis. One of them, reported by three independent groups, is on chromosome 16; the other is at 20p.

We report a Caucasian Brazilian family with 26 members distributed in three generations affected by a late onset autosomal dominant motor neurone disease. Clinical and neurological examination of 11 living members was compatible with the diagnosis of Caucasian amyotrophic lateral sclerosis and motor neurone disease and long survival.

METHODS

Patients

Figure 1 shows the family pedigree. The probands—three affected sisters (IV-12, IV-13, and IV-14)—were referred to the Human Genome Research Center at the Department of Biology, University of São Paulo, with a diagnosis of motor neurone disease. Extended pedigree analysis showed 26 members were affected (10 men and 16 women); 15 of these already were deceased. Family members reported that patients III-26, IV-10, IV-12, IV-31, and IV-38 died of respiratory failure, although no postmortem confirmation of amyotrophic lateral sclerosis was done. The mean age of death was 49.8 (SD 8.1) years, and the mean age at onset was 38 (SD 6) years.

The diagnosis of amyotrophic lateral sclerosis was based on El Escorial revised criteria. All studies were performed after death was 49.8 (SD 8.1) years, and the mean age at onset was 38 (SD 6) years.

Key points

- Motor neurone disease includes a heterogeneous group of disorders with motor neurone involvement, such as amyotrophic lateral sclerosis, progressive muscular atrophy, progressive bulbar palsy, and primary lateral sclerosis.
- Amyotrophic lateral sclerosis is the most common adult onset form of motor neurone disease and involves lower and upper motor neurones. It is characterised by progressive muscle weakness and atrophy, with fasciculations associated with hyperreflexia and spasticity.
- One proposed mechanism for amyotrophic lateral sclerosis is degeneration of the motor neurone because of abnormal levels of toxic products that accumulate in cells. Death usually occurs by respiratory failure about 2–3 years after the first symptoms.
- About 10% of cases are familial amyotrophic lateral sclerosis, and several loci have been associated with this condition. To date, the only two genes identified have been the zinc–copper superoxide dismutase 1 (SOD1) gene, which is located on chromosome 21 (ALS1, MIM105400), and the Alsin gene, which is located at 2q33 (ALS2, MIM 205100).
- Autosomal dominant forms of amyotrophic lateral sclerosis also have been linked to 18q21 (ALS3, MIM 606640), 9q34 (ALS4, MIM 602433), and 15q15.1–q21.1 (ALS5, MIM 602099), and amyotrophic lateral sclerosis–frontotemporal dementia (MIM 105550) have been linked to 9q21-22.
- Mutations in Dynein are associated with motor neurone degeneration and defects in retrograde transport. This gene acts in the cellular division, trafficking, and transport of several proteins, such as SOD1.
- We report a large Brazilian Caucasian family with clinical and neurological signs compatible with the diagnosis of amyotrophic lateral sclerosis with slow progression.
- The disease seems to affect both sexes equally, with no evidence of clinical anticipation. Clinical onset occurs between age 31 and 45 years, and the cause of death is respiratory failure. Overall, 12 family members were examined personally. All patients had lower motor neurone symptoms, and five also had bulbar involvement. Electromyography, as well as muscle biopsies, showed a neurogenic pattern.
- We mapped a novel locus for autosomal dominant late onset amyotrophic lateral sclerosis/motor neurone disease (ALS/MND) variant at 20q13.33. The identification of a new gene for ALS/MND will contribute to our understanding of this intriguing disorder.
informed consent. Demographic and clinical data were taken and followed by a complete clinical and neurological examination. Serum levels of creatine kinase, magnetic resonance imaging, nerve conduction study (motor and sensory), and electromyography were performed in the standard way. Muscle biopsies were taken from the biceps of three affected patients (IV-7, IV-14, and IV-37).

We obtained DNA from the peripheral blood of 25 family members (11 affected and 14 unaffected) according to standard procedures. As the disorder has late onset, we included in the linkage analysis only affected patients, older unaffected siblings, and unrelated unaffected spouses. We performed haplotype reconstruction, on the basis of haplotype analysis of members from generation IV, for three patients (III-12, III-20, and III-22), as well as for member III-19.

Linkage analysis

We excluded all previously known loci for amyotrophic lateral sclerosis. We performed scan analysis with distant markers about 10 cM in length from the ABI Prism Linkage Mapping Set kit (version 2; Applied Biosystems, Foster City, CA, USA). Additional polymorphic markers were included in this analysis to refine the region. The PCR products were analysed in MegaBace 1000 DNA Sequencers (Amersham Biosciences, Little Chalfont, UK). The order of the markers was based on different genetic maps of the Marshfield Medical Research Foundation database and the National Center for Biotechnology Information (NCBI) database. All information about sequence tagged sites was obtained from UniSTS of NCBI and the genome database website. As the allele frequency varies according to the population, we analysed at least 30 chromosomes from normal Brazilian controls. For linkage analysis, we used the MLink program of the Fastlink package (Columbia University, New York, NY, USA) for two point analysis, and we assumed autosomal dominant inheritance with penetrance 1 until age 45 years (as the onset in all affected patients occurs before age 45 years), equal male and female recombination rates, and a gene frequency of 0.0001 for parametric analysis. Multipoint analysis was performed with SimWalk (version 2.86).16-18

RESULTS

Clinical features

Table 1 shows the affected family members’ clinical and neurological features, as well as the results of complementary examinations.

All patients had lower motor neurone symptoms, with signs in the four limbs, and one also had upper neurone signs. Five patients (IV-23, IV-32, IV-33, IV-34, and IV-37) also had bulbar involvement. Postural tremor, one of the first symptoms to appear, was seen in eight patients, but it was stable during disease progression (IV-1, IV-16, IV-17, IV-23, IV-32, IV-33, IV-34, and IV-37).

The symptom of painful cramps was prominent, long standing, and easily obtained, and had a disabling pattern. As lower motor neurone findings confirmed by electrophysiological studies were the most conspicuous and uniform symptom in all patients, such patients first were classified as having motor neurone disease. The presence of pyramidal signs in one patient (IV-1), who could be classified as having clinically probable, laboratory supported amyotrophic lateral sclerosis, subsequently directed the investigation to the amyotrophic lateral sclerosis/motor neurone disease (AML/MND) group.

Serum creatinine kinase was normal or slightly elevated. Electromyography in eight patients showed a neurogenic pattern compatible with motor neurone disease. Needle electromyography showed abnormal spontaneous activity such as fasciculations, fibrillations, and positive waves in the tongue and muscles of the upper and lower limbs. On effort, polyphasic and giant motor unit action potentials with reduced recruitment were seen. Sensory and motor nerve conduction studies were normal.

Analyses of muscle biopsies showed a neurogenic pattern, with groups of large and small angulated fibres and fibre type grouping.
### Table 1  Clinical and neurological features of patients affected by amyotrophic lateral sclerosis and motor neurone disease

<table>
<thead>
<tr>
<th>Patient</th>
<th>Sex</th>
<th>First symptoms</th>
<th>Age (years)</th>
<th>Test result</th>
<th>Cause of death</th>
<th>Age at death (years)</th>
</tr>
</thead>
<tbody>
<tr>
<td>IV-1</td>
<td>Man</td>
<td>Cramps</td>
<td>31</td>
<td>20.0 and 23.0*</td>
<td>Motor neurone disease</td>
<td>NA</td>
</tr>
<tr>
<td>IV-2</td>
<td>Man</td>
<td>Fasciculation</td>
<td>40</td>
<td>7.0</td>
<td>Respiratory failure</td>
<td>49</td>
</tr>
<tr>
<td>IV-3</td>
<td>Man</td>
<td>Fasciculation</td>
<td>43</td>
<td>19.3 and 28.0*</td>
<td>Motor neurone disease</td>
<td>NA</td>
</tr>
<tr>
<td>IV-4</td>
<td>Man</td>
<td>Fasciculation</td>
<td>45</td>
<td>58.0 and 61.0*</td>
<td>Motor neurone disease</td>
<td>NA</td>
</tr>
<tr>
<td>IV-5</td>
<td>Man</td>
<td>Fasciculation</td>
<td>41</td>
<td>ND</td>
<td>NA</td>
<td>NA</td>
</tr>
<tr>
<td>IV-6</td>
<td>Man</td>
<td>Cramps and tremor</td>
<td>30</td>
<td>ND</td>
<td>NA</td>
<td>NA</td>
</tr>
<tr>
<td>IV-7</td>
<td>Man</td>
<td>Weakness</td>
<td>42</td>
<td>ND</td>
<td>Motor neurone disease</td>
<td>NA</td>
</tr>
<tr>
<td>IV-8</td>
<td>Man</td>
<td>Cramps</td>
<td>25</td>
<td>18.5</td>
<td>Motor neurone disease</td>
<td>NA</td>
</tr>
<tr>
<td>IV-9</td>
<td>Man</td>
<td>Tremor</td>
<td>41</td>
<td>25.3</td>
<td>Motor neurone disease</td>
<td>NA</td>
</tr>
<tr>
<td>IV-10</td>
<td>Man</td>
<td>Tremor</td>
<td>44</td>
<td>20.3</td>
<td>Motor neurone disease</td>
<td>NA</td>
</tr>
<tr>
<td>IV-11</td>
<td>Man</td>
<td>Fatigue</td>
<td>37</td>
<td>14.0</td>
<td>Motor neurone disease</td>
<td>NA</td>
</tr>
<tr>
<td>IV-12</td>
<td>Man</td>
<td>Cramps</td>
<td>37</td>
<td>18.0</td>
<td>Motor neurone disease</td>
<td>NA</td>
</tr>
</tbody>
</table>

*Normal values: <12 SU for women and <20 SU for men.
†Signs of lower motor neurone problems: weakness, atrophy, hypotonia, hypoactive or absent reflexes, and fasciculations.

### Table 2  Results of multipoint and two point LOD score analyses for informative markers at 20q13. It is considered informative if LOD >3.0.

<table>
<thead>
<tr>
<th>Marker</th>
<th>Marker position (cM)*</th>
<th>LOD score</th>
<th>Two point at θ =</th>
</tr>
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<tbody>
<tr>
<td></td>
<td></td>
<td>Multipoint</td>
<td>0</td>
</tr>
<tr>
<td>D20s1148</td>
<td>–6.601</td>
<td>–∞</td>
<td>0.668</td>
</tr>
<tr>
<td>D20s430</td>
<td>55.60</td>
<td>2.660</td>
<td>2.301</td>
</tr>
<tr>
<td>D20s164</td>
<td>57.69</td>
<td>7.450</td>
<td>5.704</td>
</tr>
<tr>
<td>D20s94</td>
<td>58.13</td>
<td>7.417</td>
<td>5.704</td>
</tr>
<tr>
<td>D20s171</td>
<td>59.51</td>
<td>3.339</td>
<td>2.316</td>
</tr>
</tbody>
</table>

*Inferred based on data from MapView-NCBI and Ensembl website.
Fig 2 Multipoint LOD score analysis for 20q13 markers based on the Simwalk program. The most significant polymorphic markers are shown.

Linkage data
We detected a significant linkage at region 20q13.3 with the flanking markers D20S178, D20S196, D20S120, D20S100, D20S102, D20S171, and D20S173. Recombination events in two patients (IV-1 and IV-34), as well as the analysis of 12 additional polymorphic markers (D20S857, D20S1083, D20S839, D20S833, D20S606, D20S183, D20S840, D20S211, D20S1148, D20S430, D20S164, and D20S93), allowed us to locate the candidate gene in a region of about 2.7 Mb between the markers D20S430 and D20S173. According to NCBI MapView, this region contains 17 known genes, one pseudogene, and six predicted genes. Table 2 shows the logarithm of odds (LOD) scores. Two point LOD score analysis showed LOD scores >3 for several markers, with a maximum value of 6.02 at 0 = 0.0 for the marker D20S171, while multipoint analysis showed a maximum LOD score of 7.45 for the marker D20S164 (fig 2).

We are currently screening functional candidate genes in the region. Until now, the tubulin beta-1 (TUBB-1), cathepsin Z (CTSZ), and ATP synthase epsilon subunit (ATP5E) genes were screened through single strand conformation polymorphism followed by direct sequencing of all amplicons, but no pathogenic mutation was identified.

DISCUSSION
We have identified a novel locus for an autosomal dominant late onset ALS/MND at 20q13.33 in a large Brazilian Caucasian family. Recently, a new amyotrophic lateral sclerosis locus was also mapped on chromosome 20.24 This locus is located in the short arm of chromosome 20 (at 20p13), however, while ours is in the long arm.

The disease seems to affect both sexes equally, with no evidence of clinical anticipation. Clinical onset in affected patients occurs earlier than in familial amyotrophic lateral sclerosis or classic amyotrophic lateral sclerosis (mean age of 38 (SD 6) years v 46 years and 56 years, respectively). In a previous study of Brazilian patients with sporadic amyotrophic lateral sclerosis, the mean age of onset was 52 years, although the first symptoms presented before age 40 years in 18.1% of patients.25 The progression of the disease in patients from the present family, however, was slower than in classic amyotrophic lateral sclerosis. In addition, other signs, such as postural tremor and disabling cramps, are atypical, although postural tremor because of increased physiological tremor may be seen in patients with weakness.26 On the other hand, the presence of atypical signs has prompted us to search for a differential diagnosis from other causes of motor neurone disease associated with postural tremor, such as extrapyramidal degeneration, distal hereditary motor neuropathy, adult onset spinal muscular atrophy, or even an occasional association of motor neurone disease and essential tremor.

Sporadic cases of motor neurone disease with tremor or choreiform movements, or both, have been described.22 These patients, however, did not show the classical findings of amyotrophic lateral sclerosis—dementia—parkinsonism, which are typical of Guam complex. In these cases, autopsy studies showed degeneration in the lower motor neurones and neostriato—pallido—nigral system or in the upper and lower motor neurones, pallido—luyiso—nigral system, and brainstem tegmentum.23

The diagnosis of distal hereditary motor neuropathy was excluded because none of our patients had pes cavus or nerve conduction abnormalities.

Many years ago, we reported a Brazilian family with seven members affected by late onset autosomal dominant spinal muscular atrophy and symptoms that overlapped with amyotrophic lateral sclerosis.24 In this family, however, all patients showed rapid progression, with death occurring 2–3 years after onset.

We propose classifying the present family as having late onset amyotrophic lateral sclerosis/motor neurone disease with atypical signs. Recently, a family with atypical autosomal dominant amyotrophic lateral sclerosis with normal life expectancy, absence of bulbar involvement, and symmetrical distal distribution of atrophy and weakness was mapped at 9q34.2 The authors suggested that this form and distal hereditary motor neuropathy with pyramidal signs could be the same disorder. We agree with those authors’ comment that it is not easy to classify these neuromuscular conditions on the basis of clinical and neurological signs.

On the other hand, genetic findings and clinical heterogeneity with atypical findings have been common observations, and other families with this intriguing phenotype may exist. Two large Brazilian genealogies with 80 affected members with late onset (mean age 48.8 years) spinal muscular atrophy (MIM 182980) were reported several years ago.23 Patients from these families showed slow loss of muscle strength, with progressive proximal atrophy, hypotonic or absent deep tendon reflexes, and fasciculations. It will be of interest to verify if they have the disorder we describe.

Intrafamilial clinical variability in patients who carry the same pathogenic mutation has been reported, however, for many gene related diseases, such as autosomal recessive limb girdle muscular dystrophies.26 Furthermore, recent studies showed that modifier genes also could alter clinical phenotype. Faster progression of disease has been reported in patients with a mutation in the ciliary neurotrophic factor (CNTF) gene associated with the SOD1 gene compared than in those who lack the CNTF mutation.27 The authors concluded that CNTF is a modifier gene that could modulate the progression of the disease, but this finding was not confirmed by other investigators.28

On the other hand, the vascular endothelial growth factor (VEGF) gene is considered a risk factor for sporadic amyotrophic lateral sclerosis. Patients homozygous for −2578A/−1154A−1534G or −2578A/−1154G−614G in the VEGF promoter/leader sequence were shown to be at 1.8 times greater risk of amyotrophic lateral sclerosis.29

Conclusion
We mapped a novel gene for an ALS/MND variant at 20q13.33 and are currently screening functional candidate genes in the region for mutations. According to the NCBI Map Viewer, the flanking markers D20S430–D20S173 span a region of about 2.7 Mb that contains 17 known genes, six predicted genes, and one pseudogene.

The TUBB-1 gene was considered to be a strong candidate gene, because it is the major component of microtubules and, recently, alterations of the axonal transport and microtubule
network have been shown to be potential causes of motor
development in amyotrophic lateral sclerosis model mice.16 No mutation was found, however, in this gene or in the
CTSZ or ATP5E genes, although the possibility of an
alternative splicing or other post-transcriptional mechanism has
not been ruled out yet.

Identification of the causative gene will be very important
for enhancing our understanding of the underlying patholo-
gical mechanisms in this heterogeneous group of disorders and hopefully will contribute to the opening of new avenues for future therapies.

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CORRECTION

Faivre L, Cormier-Daire V, Lapierre JM, et al. Deletion of the SIM1 gene (6q16.2) in a patient with a Prader-Willi-like phenotype (J Med Genet 2002;39:594–96). An erratum has been detected in the reverse primer sequence used for amplification of the intragenic microsatellite polymorphic marker of the SIM1 gene. The sequence should read 5’-CTCTCCCTCGCTGCTGATC-3’ instead of 5’-GATCAGCAGGCAGGAGAG-3’.