Clinical and molecular analyses of a Chinese spinocerebellar ataxia type 7 family that includes infantile-onset cases

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Abstract

Background: Spinocerebellar ataxia type 7 (SCA7) is a rare subtype of SCA in the Chinese population. To the best of our knowledge, no Chinese infantile-onset cases confirmed by molecular analysis have been reported. Methods: Clinical and molecular analyses were performed in a Chinese family with several members clinically diagnosed with SCA7. Results: After molecular analysis of ATXN7, the father and his daughter were shown by the agarose gel electrophoresis analysis to have one expanded allele and one normal allele. DNA sequencing showed the diplotype CAG repeats in the father and daughter to be 50/11 and 189/10, respectively. Based on this result and observed clinical features, the daughter was diagnosed with an infantile-onset SCA7. Conclusion: This is the first report of infantile-onset SCA7 in the Chinese population confirmed by molecular analysis. Our data also indicate that life expectancy is longer for infantile-onset cases without multi-organ damage, compared to cases with multi-organ damage.

INTRODUCTION

Spinocerebellar ataxia type 7 (SCA7) is associated with a variety of clinical manifestations, including progressive cerebellar ataxia, dysarthria and decreased visual acuity. Compared to other SCAs, impairment of color vision and the presence of pigmentary maculopathy are distinguishing clinical features of SCA7 patients. SCA7 is caused by expansions of CAG repeats located in exon 3 of the ATXN7 gene on chromosome 3p14-21.1 The number of CAG repeats is within the range of 4-35 in normal individuals and 38-406 in SCA7 patients. In patients, the age of onset inversely correlates with the number of CAG repeats. Rare infantile-onset cases (defined as onset age less than 2 years) are characterized by CAG repeat numbers over 130.1 The frequency of SCA7 varies among ethnic groups. SCA7 is the most common subtype of SCAs in Sweden and Finland, but is rare in China. To our knowledge, only 8 SCA7 families and 1 sporadic SCA7 case have previously been reported for mainland Chinese. No molecularly confirmed infantile-onset case, however, has been previously reported. Here, we report the first Chinese infantile-onset SCA7 case confirmed by molecular analysis.

METHODS

Subjects

The pedigree under study is shown in Figure 1. The original proband (III28) was clinically diagnosed with SCA7 according to previously published standards by two senior neurologists in March 2008 and followed for more than 3 years. Information on family history and pedigree was obtained through personal interviews with III28 and III30. Genomic DNA of III28, III30 and IV25 was extracted from (EDTA-treated) peripheral blood using a TIANamp Blood DNA Kit (TIANGEN, Beijing, China). Informed consent was obtained from III28 and III30 and informed consent for IV25 was obtained from her guardian, III28. The protocol was approved by the Ethics Committee of Huashan Hospital.
The CAG repeat expansion located in exon 3 of ATXN7 was amplified using SCA7-F1/SCA7-R1 primers as described previously. PCR amplifications were performed in a total volume of 25 μL containing 0.10 μg of genomic DNA, 0.10 μmol/L of each primer, 100 μmol/L of each dNTP and 1.25 units of LA Taq polymerase with 12.5 μL 2x GC buffer I (TaKaRa, Chiba, Japan), using a T-Gradient Thermoblock PCR system (Biometra, Gottingen, Germany). After an initial denaturation for 2 minutes at 94°C, the PCR reaction mixes underwent 30 cycles of denaturation at 94°C for 30 seconds, annealing at 52°C for 45 seconds and extension at 72°C for 1 minute, followed by a final extension at 72°C for 5 minutes. The PCR products were electrophoresed in a 2.5% agarose gel, with DL2000 (TIANGEN, Beijing, China) and 100bp DNA Ladder (TIANGEN, Beijing, China) used as DNA size markers. The PCR products were purified and sequenced as previously described.

RESULTS

Clinical features

Proband III28, a 27-year-old male, first recognized his visual impairment when he was 15 years old. Along with progressive visual dysfunction, he gradually developed impairment of color vision in the red-green axis, staring gaze, dysarthria, dysphagia, and impairments in hearing and memory. He did not complain of gait disturbances, however, until his last follow-up (June 2011). Physical examinations showed ophthalmoplegia, hyperactive deep tendon reflexes and bilateral Babinski signs. Ophthalmological examinations showed a small increase in latency of P100 in the visual evoked potential test (VEP) showed and a normal electroretinogram (ERG). Fundi of both eyes were also normal (Figure 2). Magnetic resonance imaging (MRI), however, revealed minor regions of atrophy in the brain stem and cerebellum. (Figure 3A).

IV25 was 30 months old when she first came to medical attention in March 2008. She was normal with respect to birth and early developmental milestones, including head raising, rolling-over, crawling and sitting. She had difficulties, however, learning to walk when she was 10 months old and, at present, can still not walk independently. She developed hypotonia with head lag by 12 months, at which time her vision was also significantly impaired. When examined at 30 months, she presented with dysphagia, and could not speak in sentences: her speech being restricted to simple words like “mama” and “papa”. MRI scans revealed regions of atrophy in the brain stem and cerebellum (Figure 3B). Both electrocardiogram (ECG) and echocardiography were normal. Her father (III28) refused to allow ophthalmological examinations. Her symptoms worsened gradually and she died at an age of 37 months.
Figure 3. Sagittal T1-weighted brain MRI showing mild and severe pontine and cerebellum atrophy in III28 (A) and IV25 (B), respectively.

III30, the younger brother of III28, did not show any abnormalities and was normal in general neurological examinations. Individual III8 presented with progressive visual dysfunctions and difficulty in walking when she was a teenager, and, at present, cannot walk without aid. IV7, the son of III8, was born with severe generalized hypotonia, and died at an age of 6 months.

Genetic Analysis

We were unable to contact many of the family members still thought to be alive, and several others declined to participate in our study. For this reason, genetic investigations were performed only for family members III28, III30 and IV25. Individuals III28 and IV25 were shown to have one expanded allele and one normal allele by the agarose gel electrophoresis analysis (Figure 4). After sequencing, the copy numbers of CAG repeats were 50/11 in III28 (Figure 5) and 189/10 in IV25 (Figure 6).

DISCUSSION

SCAs comprise a heterogeneous group of neurodegenerative disorders characterized by progressive cerebellar ataxia, dysphagia, and dysarthria. Decreased visual acuity and pigmentary maculopathy, however, seem to be characteristic features of SCA7. The CAG repeat number of SCA7 is inversely correlated with onset age,
similar to other CAG repeat expansion disorders, including SCA1, SCA2, SCA3, Huntington disease (HD) and spinal bulbar muscular atrophy (SBMA). Expansions longer than 70 CAG repeats typically result in onset of SCA7 before 10 years of age, while repeats longer than 130 produce infantile-onset cases.\(^3\)

To our knowledge, only 8 infantile-onset SCA7 cases confirmed by molecular analysis of \(ATXN7\) have been reported.\(^{13-18}\) These cases displayed several unique features (Table 1). The youngest case reported in the Chinese population was 4 years old at onset and carried 97 CAG repeats.\(^{11}\)

In addition, Hsieh and coworkers reported an infantile SCA7 case in the Taiwanese population that was confirmed by PCR-based Southern blot analysis, but not analyzed for the number of CAG repeats.\(^{19}\) In the current study, IV25 was shown to carry an \(ATXN7\) allele with 189 CAG repeats. She had difficulty in learning to walk when she was 10 months old and developed hypotonia with head lag by the time she was 12 months old. She was diagnosed with infantile SCA7 and is the first infantile SCA7 case confirmed by analysis of the number of CAG repeats in the Chinese population.

Figure 6. Chromatogram of IV25 carrying normal allele with 10 CAG repeats and expanded allele with 189 CAG repeats.
Table 1: Features of infantile SCA7 cases ascertained by molecular analysis

<table>
<thead>
<tr>
<th>No.</th>
<th>Clinical features</th>
<th>Onset/death age(month)</th>
<th>CAG repeat</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Failure to thrive, PDA, hypotonia, unable to roll or sit, poor oral control, possible pigment granularity, abnormal renal function, cerebellar and brainstem atrophy, died of multi-organ failure.</td>
<td>7/11</td>
<td>240</td>
<td>13</td>
</tr>
<tr>
<td>2</td>
<td>Generalized limb tremor, delayed developmental milestones, dysphagia, pigmentary degeneration, nystagmus, generalised hypotonia, cerebellar ataxia, cerebellar and brainstem atrophy.</td>
<td>9/29</td>
<td>180</td>
<td>14</td>
</tr>
<tr>
<td>3</td>
<td>Respiratory distress, ASD, PDA, hepatomegaly, multiple hemangiomas, generalised hypotonia, CLS, retinitis pigmentosa, poor eye contact, died of multi-organ failure.</td>
<td>Birth/8</td>
<td>460</td>
<td>15</td>
</tr>
<tr>
<td>4</td>
<td>Persisting vomiting and failure to thrive, tachypnea, hepatomegaly, generalised hypotonia, ASD, PDA, retinitis pigmentosa, cerebellar atrophy, CLS, died of multi-organ failure.</td>
<td>3/5</td>
<td>325</td>
<td>15</td>
</tr>
<tr>
<td>5</td>
<td>No document</td>
<td>About 12/No document</td>
<td>220</td>
<td>16</td>
</tr>
<tr>
<td>6</td>
<td>No document</td>
<td>About 24/No document</td>
<td>180</td>
<td>16</td>
</tr>
<tr>
<td>7</td>
<td>PDA, hypotonia, acquired microcephaly, absent deep tendon reflexes, ankle clonus, hypopharyngeal dysmotility with aspiration, cerebellar atrophy.</td>
<td>2/6</td>
<td>306</td>
<td>17</td>
</tr>
<tr>
<td>8</td>
<td>Poor weight gain, PDA successfully ligated, breathing difficulties, metabolic acidosis.</td>
<td>3/7</td>
<td>230-300</td>
<td>18</td>
</tr>
</tbody>
</table>

ASD: atrial septum defect; PDA: patent ductus arteriosus; CLS: capillary leak syndrome.

Several unique features of infantile SCA7 cases compared to the child-onset and adult-onset cases are listed in Table 1. First, all infantile SCA7 cases carried more than 180 CAG repeats. Second, most had (multiple) clinical features, including hypotonia and multi-organ damage such as patent ductus arteriosus, respiratory distress, capillary leak syndrome, abnormal renal function and hepatomegaly; three died from multi-organ failure. Third, the duration of the disease duration was very short, in most cases less than 8 months. Fourth, the inheritance mode was paternal transmission in all cases, i.e., the long CAG repeats carried by infantile SCA7 cases were all derived from the fathers’ expanded CAG repeats.

In the current study, IV25 was shown to carry an ATXN7 allele with 189 CAG repeats and presented with hypotonia. She did not, however, have multi-organ damage, possible explaining why she died at age 37 months, while all cases listed in Table 1, except case 2, died before 12 months. Case 2 also did not have multi-organ damage, and the disease duration was 20 months, much longer than any of the cases with multi-organ damage. Based on these observations, we infer that multi-organ damage is present in most infant SCA7 cases and may significantly reduce survival time.
Consistent with all of the cases listed in Table 1, IV25’s greatly expanded ATXN7 allele was also transmitted from a father with an expanded ATXN7 allele. Thus, reported large expansions that produce infantile-onset SCA7 consistently show paternal transmission. In contrast to these results, however, the inheritance mode in the additional infantile SCA7 case in the current study, IV7, was apparently maternal transmission. Though we could not confirm this by molecular analysis, we could propose that IV7 is an infantile SCA7 case based on clinical features. This unusual inheritance mode, however, needs to be confirmed in other populations.

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DISCLOSURE

Conflicting Interests: None

REFERENCES