Homozygous spinocerebellar ataxia type 3 in China: a case report

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Abstract
Spinocerebellar ataxia type 3 (SCA3) is a neurodegenerative disease caused by a heterozygous CAG repeat expansion in the ataxin 3 gene (ATXN3). However, patients with homozygous SCA3 carrying expanded CAG repeats in both alleles of ATXN3 are extremely rare. Herein, we present a case of a 50-year-old female who had homozygous SCA3 with expansion of 62/62 repeats. Segregation analysis of the patient’s family showed both a contraction pattern of CAG repeat length and stable transmission. The present case demonstrated an earlier onset and more severe clinical phenotype than that seen in heterozygous individuals, suggesting that the gene dosage enhances disease severity.

Keywords
Spinocerebellar ataxia type 3, homozygous, clinical feature, repeat instability, gene dosage, stable transmission

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Introduction
Spinocerebellar ataxia type 3 (SCA3) is an autosomal dominant neurodegenerative disorder caused by the abnormal expansion of cytosine–adenine–guanine (CAG) repeats in exon 10 of the ataxin 3 gene (ATXN3). SCA3 is characterized by multiple neurological features, including progressive cerebellar ataxia, external...
ophthalmoplegia and pyramidal signs, and other manifestations such as limb spasticity, dystonia, bradykinesia, and peripheral neuropathy. Normal ATXN3 CAG repeats in healthy individuals range from 12 to 44, while pathological allele expansions in patients with SCA3 fluctuate between 52 to 87. An inverse relationship between the age at onset (AAO) and the number of CAG repeats in the pathogenic allele has been widely reported.

To date, few studies have recorded homozygous cases among patients with SCA3. Homozygous individuals carrying both expanded ATXN3 alleles present with an earlier AAO and more severe clinical symptoms than heterozygotes, which may result from an effect of gene dosage. The intergenerational CAG repeat instability can explain the clinical phenotypes more generally, however, the repeat instability of homozygous individuals’ transmission is rarely reported, with only one known case carrying all expanded CAG repeats of uniform length.

Here, we report a patient with homozygous SCA3 and a history of consanguinity. This patient exhibited expanded CAG repeats which showed a pattern of contraction and uniform transmission. Our findings provide additional clinical and genetic features for homozygous SCA3.

Case presentation

A 50-year-old female patient suffered from the slow progression of gait disturbances since the age of 37. In the following year, she began to fall frequently, which required her to use mobility aids when walking. She had complained of slurred speech since the age of 45. Symptoms such as upper limb clumsiness, dysphagia, and sleep disorders had appeared successively from the age of 46, and she is now wheelchair-bound. Her neurological examination revealed severe truncal and limb ataxia, moderate gaze-evoked horizontal nystagmus, a significantly poor finger–nose test, and heel–kneel–shin ataxia. Muscle strength and volume of the four limbs were normal, but her muscle tone was increased. Tendon areflexia and a bilateral Babinski sign were also observed. The scale for the assessment and rating of ataxia (SARA) and international cooperative ataxia rating scale (ICARS) were 28/40 and 52/100, respectively. Routine blood and urine tests were normal.

The patient was a child of a consanguineous marriage, and her parents were first cousins (Figure 1a). Her maternal grandmother and paternal grandmother were sisters, but neither had any cerebellar clinical symptoms. Her mother (III-6) developed slight gait unsteadiness at the age of 70. She is 73 years old now and has no slurred speech or dysphagia. The patient’s father (III-4) shows a slightly unsteady walking pattern which began when he was 68 years old, but no slurred speech, double vision, or dysarthria symptoms were noted. Her uncle (III-2) suffered from gait problems in the sixth decade of his life and died of unknown reasons at the age of 80. Both of the patient’s younger brothers (IV-3 and IV-4) have no ataxia symptoms at present (Table 1).

After genetic counseling, written informed consent was obtained from the patient and her parents for participation in the study, and approval was provided by the Ethical Committee of the Affiliated Hospital of Hangzhou Normal University in China. Genomic DNA extraction was conducted as previously reported. Small and large bands were identified by DNA fragment analysis based on capillary electrophoresis on an ABI 3730XL DNA analyzer (Hangzhou Cred Technology, Hangzhou, China; http://www.credbio.cn). CAG repeats of the patient were visible as 62/62 (Figure 1b), while CAG repeat sizes
of her father and mother were seen as 62/18 and 65/13, respectively.

Discussion

Genetically confirmed patients with homozygous SCA3 are scarce, with only 21 cases reported in the literature before this report.\textsuperscript{3–10} We reviewed the genetic and clinical features of all 22 patients with homozygous SCA3, including our own. Most homozygote SCA3 carriers had the characteristic symptoms of progressive cerebellar ataxia, including gait or limb ataxia. Other manifestations, such as pyramidal signs, dystonia, peripheral neuropathy, cognitive impairment, psychiatric disturbance, and sleep disorders were also detected. The lengths of both expanded alleles were significantly inversely correlated with the AAO in all homozygotes, suggesting that CAG repeat expansion in SCA3 determines the AAO in a fully dominant fashion.
Repeat instability is an important feature of the intergenerational transmission of SCA. CAG repeat expansions are highly unstable in transmission from parents to offspring, and the transmission conditions of expansion, contraction, and stability account for 58.9%, 23.2%, and 17.8%, respectively, of Chinese SCA3.13 The pattern of contraction and stable CAG repeats during transmission is found in the present homozygous patient, with the maternal transmission resulting in a contraction of –3 CAG repeats, and the paternal one resulting in a uniform transmission. Notably, this phenomenon differs from a previously reported case in which paternal transmission was more unstable than maternal transmission.14 DNA methylation levels in the \textit{ATXN3} promoter are associated with (CAG)n instability in SCA3.15 Three transcription-coupled repair genes including DNA replication, repair, and recombination genes contribute to the CAG repeat instability of the SCA3.16 However, the details are not fully understood, so additional studies with more samples are necessary to further explore CAG instability in SCA3.

Although the expanded CAG repeat size was almost the same in the present family (62 for the father and patient, and 65 for the mother), the patient presented an earlier AAO and more severe clinical phenotype compared with her heterozygous parents. This suggests that the gene dosage may have a greater influence on clinical features than the size of the CAG repeat expansion, which demonstrates a loose correlation with the clinical features of SCA3. We speculate that \textit{ATXN3} haploinsufficiency or a null gene would lead to embryonic lethality, which is why no patient has been reported.10 The increasing accumulation of DNA damage and total levels of polyubiquitinated proteins have been observed in the post-mortem tissue of patients with SCA3, which supports the contribution of the loss of \textit{ATXN3} to disease.17,18

\begin{table}[h]
\centering
\caption{Summary of clinical and genetic data of affected family members.}
\begin{tabular}{|c|c|c|c|c|c|c|c|}
\hline
Case ID & Sex & Onset age (years) & Duration (years) & CAG repeats & Clinical features & SARA & ICARS \\
\hline
III-2 & male & 63 & 17 & n.a. & n.a. & n.a. & n.a. \\
III-4 & male & 68 & 4 & 18/62 & Horizontal gaze-evoked nystagmus, mild dysarthria, spastic gait disturbance & 4/40 & 8/100 \\
III-6 & female & 70 & 3 & 13/65 & Horizontal gaze-evoked nystagmus, unsteady gait & 5/40 & 10/100 \\
IV-1 & female & 37 & 13 & 62/62 & Wheel-chair bound, mild gaze-evoked horizontal nystagmus, severe dysarthria, slight dysphagia, tendon areflexia, positive Babinski’s sign & 28/40 & 52/100 \\
V-1 & female & Preclinical stage & none & n.a. & Preclinical stage & none & none \\
\hline
\end{tabular}
\end{table}

CAG, cytosine–adenine–guanine; SARA, scale for the assessment and rating of ataxia; ICARS, international cooperative ataxia rating scale; n.a., not available.
possible that homozygosity enhances the clinical phenotype through a complete loss of function of normally expressed ataxin-3, which disrupts cellular homeostasis. Further analysis of homozygous patients should be conducted to clarify the molecular mechanisms of how gene dosage affects SCA3.

Conclusions
We report one new patient with homozygous SCA3 with CAG repeat sizes of 62/62. Our patient presented with an earlier AAO and more severe clinical features than her heterozygous parents. The sizes of the expanded CAG repeat in our patient represented a relatively rare uniform and more common contraction transmission. Gene dosage may play a role in the progression of SCA3 disease.

Research ethics and patient consent
The study was approved by the Ethics Committee for Human Research in the Affiliated Hospital of Hangzhou Normal University (approval number 11). Written informed consent was obtained from the participants prior to enrollment in the study.

Availability of data and materials
The data that support the findings of this study are available on request from the corresponding author. The data are not publicly available because they contain information that could compromise research participant privacy.

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Declaration of conflicting interest
The authors declare that there is no conflict of interest.

Authors’ contributions
YCC designed the work. DL, MEW, MLZ, LNZ, JYY, MQQ, and YJ initiated the project. YCC, DL, MEW, MLZ, LNZ, JYY, MQQ, YJ, and XDL collected and analyzed the data. YCC wrote the manuscript. XDL commented and revised the manuscript. XDL supervised all aspects of the project. All authors read and approved the final manuscript.

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