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Clinical features of hereditary spinocerebellar ataxia diagnosed by molecular genetic analysis

利用分子遺傳分析診斷為遺傳性脊髓小腦共濟失調的臨床特徵

Objective. To assess the frequency and clinical features of different types of hereditary spinocerebellar ataxia in Hong Kong.
Design. Cross-sectional study using a questionnaire and clinical examination, with the majority of the information retrospectively collected.
Setting. Three regional hospitals, Hong Kong.
Participants. All patients with spinocerebellar ataxia that was confirmed by molecular genetic tests between January 2001 and October 2003.
Main outcome measures. History, latest physical examination results, clinical investigation results, and genetic profiles.
Results. A total of 16 Chinese patients had received diagnoses of spinocerebellar ataxia. These patients had spinocerebellar ataxia type 1 (n=3), spinocerebellar ataxia type 3 (Machado-Joseph disease; n=12), and dentatorubro-pallidoluysian atrophy (n=1). The most common manifestation was ataxia (15/16), followed by pyramidal signs (12/16). Other features such as bulbar dysfunction, ophthalmoplegia, neuropathy, and cognitive impairment were present but variable.
Conclusions. The clinical manifestations of different types of spinocerebellar ataxia overlap, and genetic study is necessary to confirm the diagnosis. The frequency of spinocerebellar ataxia type 3 is greater than that of other types among these Chinese patients. The age of onset of this type may correlate inversely with the number of CAG repeats.

目的：評估各類遺傳性脊髓小腦共濟失調在香港的發病率與臨床特徵。
設計：利用問卷和臨床檢查的結果進行橫斷面研究，大部分資料是病例過後收集的。
安排：三所分區醫院，香港。
參與者：在2001年1月至2003年10月期間，以分子遺傳分析確診患上脊髓小腦共濟失調的所有病人。
主要結果測量：病史、最新的身體檢查結果、臨床檢查結果，以及遺傳特徵。
結果：共有16位華裔病者接受脊髓小腦共濟失調的診斷測試，當中有3位患甲型脊髓小腦共濟失調，12位患丙型脊髓小腦共濟失調（即Machado-Joseph疾病），餘下1位患上紅色齒狀核—蒼白球萎縮症。最常見的表面徵狀是共濟失調（16位中有15位），次之為錐體標記（16位中有12位），其他散見於個別病人身上的還有延髓機能障礙、眼肌麻痺、神經病和認知損害。
結論：不同類型的脊髓小腦共濟失調，其表面徵狀可以相同，所以必須通過遺傳分析來確診。在華裔病者中，患丙型脊髓小腦共濟失調的比率相對高於其他類型，而該症發病年齡可能和CAG重複序列的數量呈負相關。

Key words:
Epidemiology;
Friedreich ataxia;
Genetic diseases, inborn;
Machado-Joseph disease;
Spinocerebellar ataxia

關鍵詞：
流行病學；
弗里德賴希共濟失調；
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Introduction

Clinical manifestation of ataxia is a common neurological presentation. Patients display uncoordinated movements and an unsteady gait, and they also have speech difficulties. By taking a history, conducting a clinical examination, and performing laboratory studies, physicians can exclude several differential diagnoses: a lesion occupying the posterior fossa/cerebellar space, alcohol dependency, hypothyroidism, vitamin E deficiency, demyelinating disease, and several metabolic disorders that manifest in adulthood.

Table 1. Patient characteristics

Patient No.	Sex/ age (years)	Duration of symptoms (years)	Age of onset (years)	Duration of walking aid used (years)	Duration of symptoms before walking aid used (years)	Spino-cerebellar ataxia type	No. of CAG repeats	Family history	Parent affected	Abnormal findings[‖]		
										MRI	EMG	EEG
1	F/61	8	53	2	6	1	47	No	U[‡]	Yes	No	No
2	F/53	10	43	1	9	1	43	No	U	Yes	No	No
3	M/33	10	23	2	8	1	52	Yes	Father	Yes	ND	ND
4	M/11	9	2	9	0	DRPLA[†]	71	Yes	Father	Yes	ND	Yes
5	M/42	12	30	5	7	3	67	Yes	Father	Yes	Yes	No
6	M/31	4	27	2	2	3	72	No	U[§]	No	No	No
7	F/44	2	42	1	1	3	69	Yes	U	No	Yes	No
8	M/39	3	36	W[*]	3	3	64	Yes	Mother	No	No	No
9	F/47	4	43	W	4	3	67	Yes	Mother	No	No	No
10	F/49	9	40	W	9	3	67	Yes	Mother	No	No	No
11	M/64	14	50	13	1	3	66	Yes	Mother	Yes	Yes	No
12	F/25	12	13	W	12	3	74	Yes	Father	No	ND	ND
13	M/38	3	35	W	3	3	59	No	U	Yes	ND	No
14	M/49	9	40	W	9	3	64	Yes	Mother	Yes	ND	ND
15	F/51	2	49	W	2	3	68	Yes	Mother	Yes	ND	ND
16	F/41	3	38	W	3	3	70	Yes	Mother	Yes	No	ND

* W able to walk independently
† DRPLA dentatorubro-pallidoluysian atrophy
‡ U unknown or both parents are unaffected
§ This patient's mother died early and two siblings had ataxia
‖ MRI denotes magnetic resonance imaging; EMG electromyography; EEG electroencephalography; ND not done

According to the Online Mendelian Inheritance in Man website,[1] more than 400 genetic diseases are associated with ataxia. Hereditary spinocerebellar ataxias (SCAs) are a group of such diseases; the prevalence of SCA is three per 100 000 in the West.[2] Because SCAs are heterogeneous neurodegenerative disorders that are characterised by late-onset ataxia and various other features, diagnosis requires assessment for the presence of ataxia and time of disease onset, a family history, examination of the clinical characteristics, and a confirmative genetic study to delineate the exact disease type.[3]

The last decade has witnessed significant progress in the diagnosis of genetic diseases; soon, definitive diagnoses will be possible for 60% to 70% of patients with hereditary ataxia, including SCA.[4] To the best of our knowledge, there has not yet been a study of SCA in our locality. In this article, we report the clinical features and genotypic findings of a series of patients with SCA in three regional hospitals in Hong Kong.

Methods

The Princess Margaret Hospital has provided genetic tests for hereditary SCA since January 2001. Polymerase chain reaction amplification of the trinucleotide (CAG) repeat region allowed detection of SCA types 1, 2, 3, 6, 7, 8, and 12, as well as dentatorubro-pallidoluysian atrophy (DRPLA).[5-9] To determine the CAG repeat size, highly denaturing polyacrylamide-gel electrophoresis (with 10% gels) was used, followed by visualisation with a gold stain (SYBR stain; Molecular Probes, Eugene, US). Direct sequencing was also performed on selected samples to confirm the sizes of repeats.

Between January 2001 and October 2003, the laboratory diagnosed SCA in 16 patients from three regional hospitals (the Princess Margaret Hospital, the Caritas Medical Centre, and the Pamela Youde Nethersole Eastern Hospital). All 16 patients were recruited into this study. The participating neurologists critically reviewed their patients and answered a standard questionnaire. Additional tests such as magnetic resonance imaging, nerve conduction study, electromyography, electroencephalography, and the mini-mental state examination were performed at the discretion of the attending neurologist. Majority of the investigation results have already been done. For some tests which were not done, patients would be asked back for the tests. For each patient, information on history (including a detailed family history), results from the physical examination, laboratory test results, and genotypic findings were tabulated and analysed. The Ethics Committee of the Kowloon West cluster gave approval prior to the survey.

Results

The 16 Chinese SCA patients came from 14 families. Twelve patients had SCA3, three had SCA1, and one had DRPLA. During the study period, a further 26 patients were referred to the hospital laboratory, but their genetic test results were negative.

Twelve of the 16 patients with SCA had a family history of ataxia. Most of the affected family members had died in China or had separated from the index patients. Hence, confirmation of SCA by genetically testing family members was not possible, except for patients 8, 9, and 10 who came from the same family and belonged to the same generation. For this reason, the stability of CAG repeats

Table 2. Clinical features of patients

Patient No.	Cerebellar signs*	Pyramidal signs†	Extrapyramidal signs‡	Ophthalmoplegia/ upward gaze palsy	Blepharospasm	Bulging eye	Brainstem signs§	Peripheral neuropathy‖	Dementia¶	Epilepsy/ myoclonic jerks	Urinary incontinence/ urgency	Walking aid needed
1	Y	Y	Y	N	N	N	Y	N	N	N	N	Y
2	Y	Y	N	N	N	N	Y	N	Y	N	N	Y
3	Y	Y	N	N	N	N	Y	Y	N	N	N	Y
4	Y	Y	Y	N	N	N	Y	N	Y	Y	N	Y
5	Y	Y	Y	Y	Y	N	Y	N	N	N	Y	Y
6	Y	Y	N	N	N	N	Y	N	N	N	N	Y
7	Y	Y	N	N	N	N	N	Y	N	N	N	Y
8	Y	N	N	N	N	N	N	N	N	N	N	N
9	Y	N	N	N	N	Y	Y	N	N	N	N	N
10	Y	Y	N	N	N	N	Y	Y	N	N	N	N
11	Y	Y	Y	N	N	N	Y	Y	Y	N	Y	Y
12	Y	Y	N	N	N	N	N	N	N	N	N	N
13	N	Y	N	N	N	N	N	N	N	N	N	N
14	Y	N	N	N	N	N	Y	N	N	N	N	N
15	Y	N	N	N	N	N	N	N	N	N	N	N
16	Y	Y	N	N	N	N	N	N	N	N	N	N

* Ataxia, tremor, dysdiadocokinesia, hypotonia, dysarthria, nystagmus, saccadic eye movement
† Hyper-reflexia, spasticity, up-going plantar response, amyotrophy
‡ Bradykinesia, dystonia, rigidity, choreoathetosis
§ Severe dysarthria, dysphagia, choking, tongue fasciculations
‖ Decreased deep tendon reflex, decreased proprioception
¶ Cognitive impairment, mini-mental state examination score of <26/30, decreased attention, reduced memory

between successive generations and the effect of parental sex on the number of CAG repeats in the next generation could not be determined.

There was an inverse correlation between the number of CAG repeats and the age of onset of symptoms in SCA3 (Fig); however, this association was statistically non-significant, probably because of the small sample size (Pearson's correlation coefficient, $r = -0.448$; P=0.144). Patient 12 had the most CAG repeats (74), had been symptomatic for 12 years, and was still ambulatory (Table 1). In contrast, patient 11 had 66 CAG repeats, his symptoms started at the age of 50 years, and after 1 year, he required a walking aid. Still, the relationship between the number of CAG repeats and the duration of symptoms before the need for a walking aid (which reflects disease severity) could not be proven in our study (not shown).

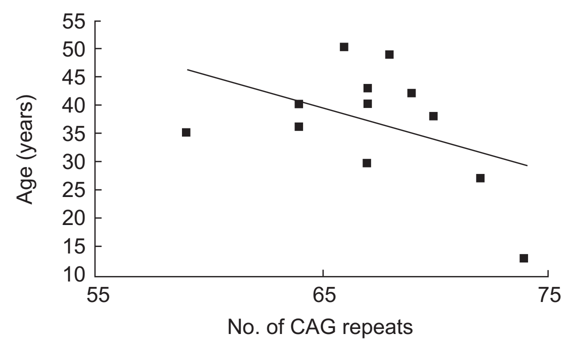

Fig. Correlation between CAG repeat number and the age of onset of spinocerebellar ataxia type 3

Although all 16 had genetic evidence of SCA, all but one patient (patient 13) had ataxia, making it the most common manifestation (Table 2); ataxia was the presenting feature for referral for these 15 patients. Pyramidal signs were the next most common presenting feature and were present in 12 patients. Eleven patients had both cerebellar ataxia and pyramidal signs. Ten patients have bulbar signs: nine showed choking; eight, severe dysarthria; seven, dysphagia; and three, tongue fasciculations (not shown). Three patients had peripheral numbness and four had an absent deep tendon reflex. One patient had ophthalmoplegia and upward gaze palsy, and another had lid retraction and blepharospasm.

Three patients had dementia, according to results of the mini-mental state examination (score of <26, on a scale of 0-30). One patient had epilepsy; no patient had psychosis. Magnetic resonance imaging scans showed cerebellar atrophy in 10 patients, brainstem atrophy in five, cerebral atrophy in six, and enlargement of the lateral ventricles in four.[10] Nerve conduction studies showed peripheral polyneuropathy in three patients (one patient with an absent deep tendon reflex did not undergo these tests). Electroencephalography showed epileptic discharges and slow waves in one patient (patient 4).

Discussion

Spinocerebellar ataxia type 3 (Machado-Joseph disease) has been reported to be one of the most prevalent types of

SCA in many Asian ethnic groups,[2-4,11] including in China, Singapore, and other parts of Asia.[12-14] In a population-based epidemiological survey from Japan, the prevalence of all types of SCA was reported to be 17.8 per 100 000.[15] However, in that survey, 20.4% of the inhabitants were older than 65 years. Another survey in Singapore showed that SCA3 was the most common type, and accounted for 53.4% of all cases of SCA.[16] We have a similar finding in this study (12/16; 75%).

Spinocerebellar ataxia type 3 is an autosomal dominant, multisystem neurodegenerative disorder that is characterised by ataxia and pyramidal signs, as well as various patterns of parkinsonism and extrapyramidal signs. Less frequent signs are external ophthalmoplegia, facial myokymia, and upward gaze palsy.[17] The disease was first described in individuals of Portuguese-Azorean descent, but was found to be one of the most common types of SCA worldwide.[18,19] The genetic defect is due to abnormal amplification of the CAG repeat at the 3' end of the coding region on the long arm of chromosome 14 (14q24.3-32.1).[20-22]

Spinocerebellar ataxia is not rare; this is also true of the other trinucleotide repeat syndromes.[23] With 16 cases diagnosed in three hospitals in our locality in less than 3 years, we expect that more cases of SCA, as well as of other hereditary neurological diseases, will be diagnosed in the future. The paucity of cases so far is probably because of a lack of awareness, expertise, and facilities.[24] When expertise and facilities are available, clinical awareness will help to increase the number of correct diagnoses. Therefore, SCA is a possible diagnosis in patients who present with ataxia and have a family history of SCA, especially when the usual search for non-genetic causes yields negative results.

Abnormal alleles in patients in our study contained CAG repeats that ranged in copy number from 59 to 74. Other studies have shown that the expanded allele and the normal allele do not overlap. There is no allele which has a number between the expanded and normal allele to cause confusion. Furthermore, there has been no example of an allele whose size is between that of a normal allele and that of an expanded allele.[25,26] We did not perform genetic tests for asymptomatic individuals.[27] Asymptomatic carriers have been described in the literature: they were relatives of SCA patients and had CAG repeats with a copy number of between 66 and 81; their ages ranged from 7 to 31 years.[10]

A positive family history supports a hereditary origin of disease. Studies based on several families have found that the number of CAG repeats increases by a mean of 0.86 to 1.60 between each successive generation.[13-15] This effect may be more prominent for paternal transmission.[14] Four of the patients in our study did not have a family history of SCA. However, the best family history patients can provide with certainty is often limited to two generations. Details about the third generation are often recalled from vague memories because of a lack of communication among family members. Hence, a negative family history should not eliminate the possibility of SCA.[7,10] Other reasons for a negative family history, such as adoption or a spontaneous mutation, must be considered.[17]

In our study, on the basis of the clinical findings, the laboratory tested for SCA3, then SCA1, and—if the suspicion for SCA was high—other types. No cases of SCA2 were found in this study. Patient 13 had a very unusual clinical course. He had rapidly progressive myelopathy with pronounced pyramidal signs. He had undergone an open-heart surgery for congenital heart disease 10 years before the onset of symptoms. Although a battery of thorough investigations, including tests for human T-cell lymphotropic virus type I antibody and HIV status, were performed, the only abnormal finding was the atrophic changes in his brainstem and cervical cord, as detected by magnetic resonance imaging. He had severe spastic tetraparesis, and it was difficult to be certain whether he had any ataxia or not. According to thorough family histories, no relatives of patients had suffered from any neurological symptoms; however, no formal clinical visit or genetic test was performed for family members. A genetic study for SCA was then performed for patient 13 despite his negative family history. This case illustrates that genetic testing is particularly helpful in making a definitive diagnosis in patients with a negative family history.[10,28]

There are several pitfalls in the diagnosis of SCA. There are several hundred causes of ataxia: several diseases, such as Parkinson's disease, Friedreich's ataxia, Huntington's disease, ataxia telangiectasia, Kennedy's disease, and multiple system atrophy, can mimic SCA.[29] So far, more than 20 SCA loci have been identified, but genetic tests are available for some of the genotypes only.[30] Clinical features alone are generally not reliable in differentiating different genotypes of SCA, except for SCA7, which has the unique feature of pigmentary maculopathy. A negative genetic test result cannot exclude SCA because the gene may not be included in the screening panel or it may not yet be discovered. However, the presence of ataxia should prompt the clinician to critically review the patient and consider an alternative diagnosis.

In conclusion, our study illustrates that SCA is an important cause of idiopathic cerebellar ataxia and myelopathy among Chinese patients. Genetic testing is necessary for selected cases to diagnose these hereditary diseases. The correct diagnosis, however, is only the beginning. The patient should be treated holistically and given psychological, psychiatric, and social support. Genetic counselling and family planning advice should also be given before the genetic tests.

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