Journal of the Neurological Sciences 354 (2015) 75–78

Contents lists available at ScienceDirect

Journal of the Neurological Sciences

journal homepage: www.elsevier.com/locate/jns

Evidence for a common founder effect amongst South African and Zambian individuals with Spinocerebellar ataxia type 7

Danielle C. Smith [a], Masharip Atadzhanov [b], Mwila Mwaba [b], Leslie Jacqueline Greenberg [a,*]

[a] Division of Human Genetics, Department of Clinical Laboratory Sciences, Institute of Infectious Disease and Molecular Medicine, Faculty of Heath Sciences, University of Cape Town, Cape Town, South Africa
[b] Department of Internal Medicine, School of Medicine, University of Zambia, Lusaka, Zambia

ARTICLE INFO

Article history:
Received 4 March 2015
Received in revised form 8 April 2015
Accepted 30 April 2015
Available online 9 May 2015

Keywords:
Spinocerebellar ataxia type 7
Polyglutamine
Haplotype
Founder effect
Africa
Neurodegeneration

ABSTRACT

Spinocerebellar ataxia type 7 (SCA7) is an inherited neurodegenerative disease caused by the expansion of a CAG repeat within the *ataxin 7* gene, leading to a pathogenic polyglutamine tract within the ataxin 7 protein. SCA7 patients suffer from progressive cerebellar ataxia and macular degeneration. SCA7 is considered to be rare, although founder effects have been reported in South Africa, Scandinavia and Mexico. The South African SCA7-associated haplotype has not been investigated in any other populations, and there have been limited reports of SCA7 patients from other African countries. Here, we describe the first two ethnic Zambian families with confirmed SCA7. Haplotype analysis showed that the South African SCA7 haplotype alleles were significantly associated with the pathogenic expansion in affected Zambian individuals, providing strong evidence for a shared founder effect between South African and Zambian SCA7 patients.

1. Introduction

Spinocerebellar ataxia type 7 (SCA7) is an inherited neurodegenerative disease caused by the expansion of a CAG repeat within the *ataxin 7* gene, beyond a pathogenic threshold of 36 repeats [1,2]. SCA7 patients suffer from selective degeneration of cerebellar Purkinje neurons and inferior olivary nuclei, regions of the brain that are involved in the control and coordination of movement [3]. Blindness occurs as a result of degeneration of the neural retina (photoreceptors and ganglion cells), which may be accompanied by deterioration of the optic tract and visual cortex [3,4].

SCA7 is one of the most frequent types of inherited ataxia in South African individuals, along with Spinocerebellar ataxia types 1 and 2 (SCA1, 2) [5,6]. Few South African individuals with Spinocerebellar ataxia type 3 and 6 (SCA3, 6) have been identified. SCA7 is considered to have a low occurrence across the globe, although higher frequencies have been reported in Scandinavia and Mexico, as a result of founder effects within those regions [7–9]. In South Africa (SA), SCA7 occurs almost exclusively within the indigenous Black African population, and a previous haplotype-based study provided evidence for a founder effect within this patient group [10]. There are limited reports of SCA7 in other African countries, however, isolated African SCA7 families have been included in larger international patient cohorts [11–14]. To date, the South African SCA7-associated haplotype has not been investigated in any additional populations. Here, we describe the first two SCA7 families from Zambia, and provide evidence to suggest that South African and Zambian SCA7 patients share a common founder haplotype.

2. Materials and methods

2.1. Patients and families

Ethical approval for the study was obtained from the University of Cape Town Human Research Ethics committee (HREC REF 460/2010, renewed annually) and the Biomedical Research Ethics Committee of the University of Zambia. Probands, affected and non affected family members were examined by a neurologist and individuals who provided DNA samples gave written informed consent. DNA samples were analysed from 13 family members (called Family A) from the city of Kitwe (ethnic Zambian, Bemba tribe), as well as 4 individuals from an additional unrelated family from the town of Mongu (Family B, ethnic Zambian, Lozi tribe). At the time of testing, five individuals from Family A and two individuals from Family B showed clinical symptoms associated with SCA7. The main phenotype in both families was characterized by gait and limb ataxia, dysarthria, visual loss, ptosis, ophthalmoparesis, pyramidal tract signs and dementia. The combination of these symptoms, with bilateral macular degeneration and cerebellar atrophy in

* Corresponding author at: Division of Human Genetics, N3.14, Wernher & Beit North Building, Institute of Infectious Disease & Molecular Medicine, University of Cape Town Faculty of Heath Sciences, Observatory 7925, South Africa. Tel.: +27 21 406 6299.
E-mail address: jacquie.greenberg@uct.ac.za (L.J. Greenberg).

http://dx.doi.org/10.1016/j.jns.2015.04.053
0022-510X/

CT/MRI was in line with Spinocerebellar ataxia 7 [1,3]. Cerebral malaria and HIV infection were ruled out as the causes of ataxia in all affected members. DNA was also analysed from 40 unaffected control (mutation negative) individuals from the same region (all of Zambian origin).

2.2. Genotyping of CAG repeat and microsatellite markers

Genomic DNA was extracted from blood plasma using the QIAmp DNA Mini Kit (Qiagen). All individuals were genotyped at four loci: the *ataxin 7* CAG repeat, an intronic AC repeat between exons 1 and 2 (called "AC1"), a SNP (rs3774729), and a microsatellite marker (D3S1287) as previously reported by Greenberg et al. [10]. The CAG repeat genotype of each patient was determined using a singleplex PCR or a triplet-primed PCR (TP PCR) assay [15]. Where TP PCR was used, the CAG repeat genotype of the individual was denoted using N to indicate an allele within the unaffected range, and E to indicate a pathogenically expanded allele. Detailed methods and allele sizes are provided in supplementary information.

2.3. Haplotype analysis

For haplotyping analysis, the phase of the alleles was inferred manually in each pedigree, and confirmed using the program SHEsis [16,17]. SHEsis was also used to calculate the estimated haplotype frequencies in the unaffected control cohort. The previously published haplotype-associated alleles for South African SCA7 patients were allele 6 for AC1, allele A for rs3774729 and allele 1 for D3S1287 [10].

3. Results

Four clinically non-symptomatic individuals were found to have CAG repeat expansions within the disease-causing range (three from Family A, one from Family B). All of the individuals with a pathogenic repeat had the South African SCA7 haplotype-associated alleles at each of the three markers (allele 6 at AC1, allele A at rs3774729, and allele 1 at D3S1287). Within the unaffected control group, allele 6 of AC1 was found in 8 individuals, whilst allele 1 of D3S1287 was not found in any of the control cohort. The A allele of rs3774729 was found at a high frequency in both groups (100% of patients and 91% of control samples). The South African SCA7 haplotype alleles for the three markers were significantly associated with the patient group ($p = 2.07E\text{-}05$), suggesting that the haplotype is associated with the disease, and is not simply at a high frequency in the tested control population (Table 1).

Together, these results provided evidence to suggest that the Zambian SCA7 patients from the two families shared the SA SCA7-associated haplotype. To substantiate these findings, the pedigree of each family was used to manually infer the phase of each allele (Fig. 1). Although no samples from complete parent–child trios were available to unequivocally determine the phase of each haplotype marker, pedigree analysis provided further evidence to support the supposition that the SA SCA7-associated haplotype was shared within both families.

Table 1
Comparison of calculated haplotype frequencies in Zambian SCA7 patients and unaffected controls. The South African SCA7 associated haplotype is in bold. Only assigned haplotypes are presented.

Haplotype (AC1, rs3774729, D3S1287)	No. of cases (freq)	No. of controls (freq)	Chi^2	Pearson's p
1 A 4	0 (0.000)	1 (0.028)	–	
2 A 2	0 (0.000)	2 (0.056)	1.116	0.290854
2 A 3	0 (0.000)	1 (0.028)	–	
3 G 2	0 (0.000)	1 (0.028)	–	
4 A 3	0 (0.000)	3 (0.083)	1.712	0.19067
4 G 2	0 (0.000)	3 (0.083)	1.712	0.19067
4 G 5	0 (0.000)	1 (0.028)	–	
5 A 4	0 (0.000)	7 (0.195)	4.414	0.035668
5 G 2	0 (0.000)	4 (0.111)	2.33	0.126892
6 A 3	1 (0.062)	7 (0.194)	2.121	0.145265
6 A 4	0 (0.000)	1 (0.028)	–	
7 A 2	0 (0.000)	2 (0.056)	1.116	0.290854
7 A 4	3 (0.188)	2 (0.055)	1.585	0.208011
7 G 3	0 (0.000)	1 (0.028)	-	
4 G 3	1 (0.062)	0 (0.000)	1.916	0.166285
5 A 2	1 (0.062)	0 (0.000)	1.916	0.166285
5 A 3	1 (0.062)	0 (0.000)	1.916	0.166285
6 A 1	**8 (0.500)**	**0 (0.000)**	**18.151**	**2.07E-05**
7 G 4	1 (0.062)	0 (0.000)	1.915	0.166344

4. Discussion

The global prevalence of SCA7 is believed to be relatively low, apart from regions with established founder effects in South Africa and Scandinavia. Although disease-associated haplotypes have been established in both the SA and Scandinavian SCA7 patient populations, only one marker is shared between the two haplotypes (D3S1287). Due to the lack of reporting of allele calling methods for this marker, it is unclear whether the South African and Scandinavian patients share the same allele at this locus. Similarly, a haplotype-based study was undertaken in Mexico in 2013, and two of the investigated markers included D3S1287 and rs3774729, which form part of the SA SCA7-associated haplotype [8]. Ninety individuals from 19 families from the Veracruz region of Mexico were genotyped, and found to share a common haplotype. Once again, due to differences in allele reporting methods between the Greenberg and García-Velázquez studies, it cannot be determined whether the Mexican and SA SCA7 patients share the same alleles at the two marker loci. Further investigation will be required to determine whether SCA7 patients from these different geographical regions have distinct genetic haplotypes, as is the case with Huntington disease [18,19].

In this study, evidence for the existence of the SA SCA7-associated haplotype in two SCA7 families Zambia is presented. A confirmed molecular diagnosis has an immediate benefit to these families, since testing can be offered to other family members, the family can be counselled with regard to the management of affected members, and risks for current and future offspring can be discussed. These haplotyping results also suggested that the South African and Zambian SCA7 families tested share a common founder. Although some background control individuals were genotyped from the Zambian population, future studies should focus on including additional unaffected individuals from the studied regions.

The Bantu people encompasses a large ethnic group distributed across central and southern Africa, and includes numerous Bantu-speaking sub-populations in Zambia and South Africa. In light of the haplotyping results presented here, it is likely that additional SCA7 families exist in neighbouring countries such as Botswana and Zimbabwe (which exist geographically between Zambia and South Africa), and other Bantu-speaking countries further north such as Angola, Tanzania and the Democratic Republic of Congo. A shared haplotype between large groups of affected individuals may be beneficial for the design of future therapeutic interventions. The RNAi-based therapy designed by Scholefield and colleagues targets the A allele of the SNP linked to the pathogenic CAG repeat in patients with the SA SCA7-associated haplotype [20,21]. Similarly, a panel of five siRNAs have been designed to target three-quarters of US and European Huntington Disease patients [22], demonstrating the value of population-based haplotype studies.

It has been acknowledged that the body of literature on neurodegenerative disorders such as SCA7 in Sub-Saharan Africa is limited, and that there have been few population-based studies [23]. Reports on the prevalence of the inherited ataxias on the African continent have been extremely scarce. Many of the existing reports in the literature are not representative of a large geographical area, but rather of isolated cases and families. Published prevalence rates in Sub-Saharan Africa are