Genetic analysis of the *CHCHD2* gene in a cohort of Chinese patients with Parkinson disease

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Abstract

CHCHD2 has been recently reported as a causative gene for autosomal dominant Parkinson disease (ADPD) in Japanese populations. Further genetic studies of *CHCHD2* in other populations are needed. Herein, we sequenced *CHCHD2* gene in 162 patients (90 from ADPD pedigrees, 72 with sporadic Parkinson disease) and 90 healthy controls in Chinese population. We observed 5 exonic variants (c.-34C>A, c.-9T>G, c.5C>T, c.*125G>A, c.*154A>G) including 1 novel variant. No pathogenic mutation was found, suggesting that *CHCHD2* mutations may be rare in Chinese ADPD patients.

Key words: Parkinson disease, autosomal dominant Parkinson disease, *CHCHD2*, Chinese population

Introduction

Parkinson disease (PD) is a common movement disorder characterized by the cardinal features of resting tremor, bradykinesia, rigidity, postural instability with a prevalence of 1–2% in general population over the age of 60 [2]. About 5–10% of the patients show an autosomal dominant or recessive mode of inheritance, which indicated that genetic factors play an important role in the mechanism of PD [10]. With the development of high-throughput methods such as exome sequencing, many PD causative genes were found. For example, autosomal dominant PD is associated with *SNCA* [16], *LRRK2* [15], *UCHL1* [9], *VPS35* [19, 21] and *EIF4G1* [1]. However, these causative genes are unable to explain all the ADPD families.

Recently, Funayama et al. identified *CHCHD2* as causative gene in Japanese populations, which increased the monogenic causative gene of ADPD [5, 17]. However, several studies in different ethnic populations showed that mutation in *CHCHD2* were rare in PD. Jansen et al. found 3 novel putative pathogenic variants in *CHCHD2* among people of western European ancestry [8]. While Kotaro Ogaki et al. and Ming Zhang et al. did not find any definite pathogenic *CHCHD2* mutations in the Caucasian multicenter study [14, 20]. Only one study identified a heterozygous variant (c.182C>T; p.Thr61Ile) in the *CHCHD2* gene in the ADPD pedigree in Chinese populations [14], and there was no pathogenic mutations found in other Chinese populations [3, 4, 11, 13].

In order to investigate the relationship between *CHCHD2* and PD in Chinese populations, we performed polymerase chain reaction (PCR) combined with DNA direct sequencing in 162 patients (90 from ADPD pedigrees, 72 with sporadic Parkinson disease) and 90 healthy controls.

Materials and methods

The study comprised 90 patients (age 63.1±9.5 years, onset 58.0±14.5 years) of PD families (70 families have 2 or more PD patients in at least 2 consecutive generations

and 20 families had at least 2 siblings with PD), 72 SPD patients and 90 healthy controls. All patients and controls were collected by the Neurology Department of the Second Affiliated Hospital, School of Medicine, Zhejiang University. PD was diagnosed using United Kingdom PD Society Brain Bank diagnostic criteria by two or more experienced neurologists [6]. Informed consent was obtained from each participant. The institutional ethics board committees approved the study.

Genomic DNA was isolated from peripheral blood of patients by standard protocols. All coding exons of the *CHCHD2* gene were amplified by PCR. The primers used for PCR amplification cover all coding regions of the *CHCHD2* gene [5]. Each PCR product was directly sequenced on an ABI 3100 automated sequencer (Applied Biosystems, Foster City, CA). DNASTar was used for sequence alignment and analysis (DNASTar, Inc., Madison, WI) by two analysts independently.

Results

No pathogenic mutation of *CHCHD2* gene was found in any of the 90 patients. We observed 5 exonic variants as shown in table1, and out of these, 4 polymorphisms (c.-34C>A, c.-9T>G, c.5C>T, c.*125G>A) have been reported, 1 case of c.*154A>G has however not reported or recorded in dbSNP 144, 1000 genomes, Exome Variant Server (EVS) or Exome Aggregation Consortium (ExAC) and was not observed in SPD patients and controls. The frequency of the 5 polymorphisms showed no significant statistically difference among ADPD, SPD and the control group (P value > 0.05).

Table 1

CHCHD2 gene variants detected in this study

Chromosome location	Accession number	Nucleotide change	Position	Variant frequency			
				Total PD	ADPD	SPD	Controls
Chr7:56174140	rs816407	c.-34C>A	5'UTR	0.086	0.100	0.069	0.078
Chr7:56174115	rs10043	c.-9T>G	5'UTR	0.086	0.100	0.069	0.078
Chr7:56174102	rs142444896	c.5C>T	Pro2Leu	0.014	0.000	0.014	0.000
Chr7:56169419	rs8406	c.*125G>A	3'UTR	0.086	0.100	0.069	0.078
Chr7:56169390	Novel	c.*154A>G	3'UTR	0.022	0.022	0.000	0.000

Accession number (rs) is given for each known *CHCHD2* polymorphism. In this study, the variant frequency has no significant difference among Total PD, ADPD, SPD and the controls (P value > 0.05)

A1

A2

Fig.1. (A1) A heterozygous polymorphism: c.*154A>G (arrow points) and (A2) wild type (arrow points)

Discussion

Gene *CHCHD2* contains 4 exons and encodes CHCHD2 protein, characterized by an N-terminal mitochondrial targeting sequence (MTS) and twin cysteine-x9-cysteine motifs which are involved in biogenesis and regulation of enzymes in the mitochondrial respiratory chain [12].

We did not note any of the putative causal variants (c.182C>T, c.434bG>A and c.300+5G>A) described by Funayama [5], suggesting that the pathogenic mutations of this gene may be rare in Chinese ADPD patients. Moreover, mutations in the *CHCHD2* gene are rare in both general population and Chinese population with predicted frequencies of less than 0.19% (6/3086) and 0.21% (2/957)[13].

In this study, 4 reported variants (c.-34C>A, c.-9T>G, c.5C>T, c.*125G>A) and a novel variants c.*154A>G were detected. It's worth noting that the 4 single nucleotide variants (c.-34C>A, c.-9T>G, c.5C>T and c.*125G>A), which were confirmed to have different frequencies between sporadic Parkinson disease and controls by Funayama et al. [5], showed no significant difference between ADPD and controls in our study. Similarly, the association observed in the Japanese populations with the common variants, was not replicated in both the Caucasian series by Kotaro Ogaki et al. and Iqbal and Toft's study [7, 14]. What's more, we observed 1 novel variant c.*154A>G in a single heterozygous state in 2 ADPD patients, which was not observed in control group. The role of the novel polymorphism c.*154A>G needs further investigation. However, the single nucleotide variant c.5C>T (Pro2Leu) was only detected in 1 of our SPD patients, which raised a heated discussion in different

ethnic populations. Most studies in Asian populations have revealed that c.5C>T in *CHCHD2* is associated with PD[4, 5, 18], except the study conducted in Taiwanese populations[3] and the studies in other ethnic populations[7], which may partly due to ethnic difference.

In our study, we did not exclude known mutations of PD gene in the 90 ADPD patients and our cohorts were relatively small. Larger-scale genetic studies in multiple ethnic populations needs to be performed to establish *CHCHD2* associations with PD.

Conclusions

Our results suggest that *CHCHD2* mutations may be rare in Chinese ADPD patients.

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Ethical standards

This study has been performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki and its later amendments.

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