International Journal of Pediatric Otorhinolaryngology 79 (2015) 745–748

Contents lists available at ScienceDirect

International Journal of Pediatric Otorhinolaryngology

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

Genetic counseling for a three-generation Chinese family with Waardenburg syndrome type II associated with a rare *SOX10* mutation

Kaitian Chen [a,1], Ling Zong [a,b,1], Yuan Zhan [a], Xuan Wu [a], Min Liu [a], Hongyan Jiang [a,*]

[a] Department of Otorhinolaryngology, The First Affiliated Hospital, Sun Yat-sen University and Institute of Otorhinolaryngology, Sun Yat-sen University, Guangzhou 510080, PR China
[b] Department of Otorhinolaryngology, The Second Affiliated Hospital of Guangzhou Medical University, Guangzhou 510260, PR China

ARTICLE INFO

Article history:
Received 22 December 2014
Received in revised form 5 March 2015
Accepted 7 March 2015
Available online 17 March 2015

Keywords:
Genetic counseling
Hearing impairment
Nonsense-mediated mRNA decay
SOX10 mutation
Waardenburg syndrome

ABSTRACT

Objective: Waardenburg syndrome is clinically and genetically heterogeneous. The *SOX10* mutation related with Waardenburg syndrome type II is rare in Chinese. This study aimed to uncover the genetic causes of Waardenburg syndrome type II in a three-generation family to improve genetic counseling.
Methods: Complete clinical and molecular evaluations were conducted in a three-generation Han Chinese family with Waardenburg syndrome type II. Targeted genetic counseling was provided to this family.
Results: We identified a rare heterozygous dominant mutation c.621C > A (p.Y207X) in *SOX10* gene in this family. The premature termination codon occurs in exon 4, 27 residues downstream of the carboxyl end of the high mobility group box. Bioinformatics prediction suggested this variant to be disease-causing, probably due to nonsense-mediated mRNA decay. Useful genetic counseling was given to the family for prenatal guidance.
Conclusion: Identification of a rare dominant heterozygous *SOX10* mutation c.621C > A in this family provided an efficient way to understand the causes of Waardenburg syndrome type II and improved genetic counseling.

1. Introduction

Waardenburg syndrome (WS) is an uncommon auditory-pigmentary defect with variable clinical manifestations involving sensorineural hearing impairment and abnormal pigmentation of the hair, iris and skin. The incidence of WS was estimated to be 1/40,000 at birth and both genders and all races are equally affected [1]. The typical syndrome is classified into four subtypes (WS1–4) according to clinical characteristics (Table 1). Additionally, WS is genetically heterogeneous. Several genes related with neural crest development are involved (*PAX3*, *SOX10*, *SNAI2*, *MITF*, *EDN3* and *EDNRB*), with *SOX10* mutations in 15% of WS2, which is distinguished by the absence of dystopia canthorum, and 50% of WS4 [2,3], which is characterized by the presence of aganglionic megacolon. The mutation spectrums of each type in different populations are variable.

The *SOX10* gene spans about 14 kb of DNA and consists of 5 exons. It encodes a 466 amino acid transcription factor containing a central high mobility group (HMG) DNA-binding domain and a C-terminal transactivation domain. *SOX10* is essential for the development of cells in the neural crest lineage, including melanocytes and enteric ganglia. To date, more than 20 mutations in *SOX10* gene, mainly arisen *de novo*, have been identified in WS4 patients [4]. However, there are very limited reports on *SOX10* mutations that were associated with WS2, and the data from Chinese patients with WS remains poor [5–9]. Prenatal genetic counseling for WS families is impeded for the lack of understanding of the causes and the unpredictability of the inheritance.

Here, we describe a rare mutation in the *SOX10* gene associated with WS2, but not WS4 in a three-generation Han Chinese pedigree. Genetic counseling was given to this family for prenatal guidance. These findings would be beneficial for the clinician's decision-making and genetic counseling for WS families.

* Corresponding author. Tel.: +86 020 87333733; fax: +86 020 87333733.
E-mail address: jhongy@mail.sysu.edu.cn (H. Jiang).
[1] These authors contributed equally to the study.

http://dx.doi.org/10.1016/j.ijporl.2015.03.006
0165-5876/

Table 1
Summary of distinguishing characteristics of WS subtypes.

Types	Main clinical features
WS1	Hearing loss + pigmentary abnormality + **dystopia canthorum (W > 1.95)**
WS2	Hearing loss + pigmentary abnormality, **without dystopia canthorum (W < 1.95)**
WS3	Hearing loss+ pigmentary abnormality + **musculoskeletal abnormalities**
WS4	Hearing loss + pigmentary abnormality + **aganglionic megacolon**

2. Patients and methods

A three-generation Han Chinese pedigree from Guangdong province in China was presented to our department. The five years old boy (III-1) was referred for molecular diagnosis and prenatal counseling for his aunt II-3, who had normal hearing and was already pregnant for five weeks (Fig. 1). The study was approved by the institutional review board of the First Affiliated Hospital, Sun Yat-sen University. Informed consent was obtained from all the participants or guardian.

Complete medical history of this family was taken and physical examinations were performed and the possibility of environmental causes was excluded. Despite uneventful pregnancy and delivery, the proband failed the newborn hearing screening after birth. Unfortunately, no further medical intervention was carried out in this child until one year old, when hearing examinations including auditory brainstem response (ABR) and auditory steady-state evoked response (ASSR) confirmed bilateral profound sensorineural hearing impairment. Temporal computed tomography (CT) identified bilateral semicircular canal malformation and Mondini dysplasia. Vestibular function and vision was unaffected, however, both his irises were blue and he had a flat nasal root and gray hair. Other signs, such as dystopia canthorum, musculoskeletal abnormalities or aganglionic megacolon were not present. Mental and physical developments were normal. Overall, auditory and pigmentary disturbance were observed in this child. The *W* index based upon the inner canthal, interpupillary, and outer canthal distances was less than 1.85, in according with WS2. He received hearing aids but achieved poor hearing and speech ability. Besides, his father (II-2) also suffered from bilateral deafness and blue irises, and died of lung carcinoma several years ago. There was no other family history of malformations, constipation, and/or mental retardation.

Blood samples were collected from the subjects except his father and grandpa, and DNA isolation was performed using a standard chloroform extraction method. All coding exons of *SOX10*, *MITF* and *PAX3* were subsequently PCR amplified and directly sequenced using an ABI 3730 Genetic Sequencer as previously described [9]. All sequences were aligned and compared with published sequences from the NCBI (*SOX10*: NM_006941.3; *MITF*: NM_000248.3; *PAX3*: NM_000438.5). Amino acid conservation alignments were applied across different mammalian *SOX10* gene families. The pathogenicity of the alteration was further confirmed by online MutationTaster prediction [10].

3. Results

The proband was diagnosed with WS2 based on clinical manifestations according to the international criteria proposed by the Waardenburg Consortium [11]. Genetic analysis identified a rare dominant *SOX10* heterozygous mutation c.621C > A (p.Y207X) [12] (Fig. 2), which had not been reported in Chinese before. The penetrance in this family could not be determined for the lack of genetic results in I-1 and II-2. Both his mother (II-1) and aunt (II-3) carried wild type nucleotide in this locus. This variant was not detected in 50 normal Chinese individuals. No *MITF* or *PAX3* gene mutations were found.

The mutation c.621C > A occur in *SOX10* gene exon 4, and encoded Tyr207, 27 residues downstream of the carboxyl end of the HMG box. The substitution causes premature termination at amino acid 207 and is likely to be nonfunctional owing to the production of truncated protein. Amino acids around the targeted regions were highly conserved across different mammalian species *SOX10* gene family (Fig. 2). Bioinformatic prediction suggests this variant to be disease-causing probably due to nonsense-mediated mRNA decay (NMD) [10]. The pathogenicity of p.Y207X was confirmed by animal modeling study [12].

Genetic counseling was given to this family. This analysis has implications for risk prediction. First, the presence of the *SOX10* c.621C > A mutation that was probably inherited from the II-2 subject, may explain the occurrence of WS in this child and his father. An autosomal dominant inheritance pattern could be inferred, giving rise to a 50% recurrent risk in the offspring of the patient. Besides, the mutation in II-2 may arise *de novo* (a new mutation carried by II-2 subject) based on the clinical manifestation in I-1 and I-2. This genotype was in accord with the majority of

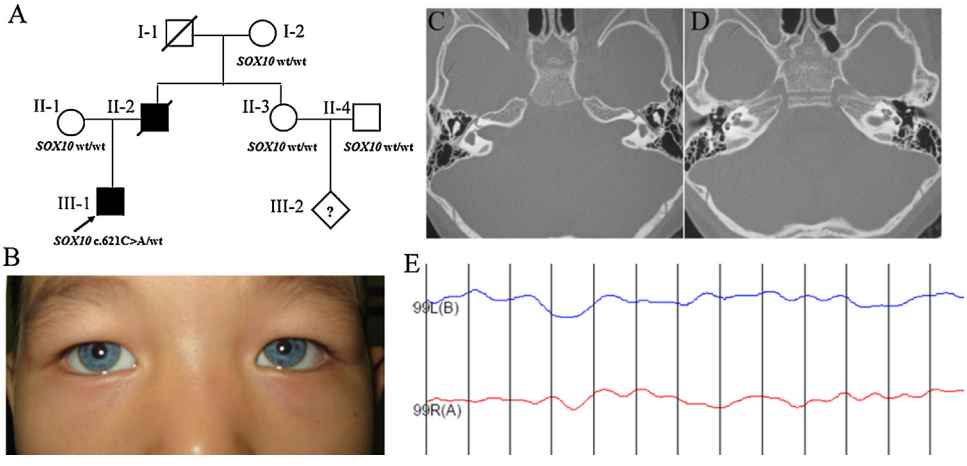

Fig. 1. The pedigree showed the heterozygous *SOX10* c.621C > A mutation (A). Clinical features of the proband included segmental iris heterochromia (B) and inner ear malformation ((C)–(D)). ABR confirmed bilateral profound deafness (E).