

CASE REPORT

CLOUSTON SYNDROME: FIRST CASE IN RUSSIA

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ABSTRACT

Hidrotic ectodermal dysplasia type 2 (HED2) or Clouston syndrome (OMIM #129500) is a rare autosomal dominant genetic disorder which affects skin and its derivatives, characterized by the major triad of features: nail dystrophy, generalized hypotrichosis, and palmoplantar hyperkeratosis. Here we describe the first case of Clouston syndrome in Russia and the molecular genetic analysis of this case.

Keywords: Clouston syndrome, Cx30, *GJB6*, Hidrotic ectodermal dysplasia type 2 (HED2).

INTRODUCTION

Hidrotic ectodermal dysplasia type 2 (HED2) or Clouston syndrome (OMIM #129500) is a rare autosomal dominant genetic disorder which affects skin and its derivatives, characterized by the major triad of features: nail dystrophy, generalized hypotrichosis with alopecia and palmoplantar hyperkeratosis, frequently accompanied by normal perspiration and dentition. The one and only genetic cause of Clouston syndrome is thought to be a mutation in the *GJB6* gene localized on chromosome 13q12 [1]. This gene encodes for a 30 kDa gap junction

protein, subunit B6 (connexin 30, Cx30), forming gap junctions between adjacent cells which allow molecules and ions to pass between them.

In some cases, mutations in the *GJB6* gene are known to cause autosomal dominant (locus DFNA3) and autosomal recessive (locus DNFB1) non syndromic sensorineural hearing loss [2,3]. It has also been reported that a mutation in the *GJB6* gene is the cause, in at least one case, of keratitis-ichthyosis-deafness (KID) syndrome [4].

Four mutations were found in Clouston syndrome patients: G11R, V37E, D50N and A88V [5]. These cases of hidrotic ectodermal dysplasia were described in British, French, Spanish, African, Malaysian and Chinese populations [6-8]. Although the cases of Clouston syndrome were reported in different populations, there are no documented cases of HED2 in Russia. Here we describe the first report of Clouston syndrome in Russia and the molecular genetic analysis of this case.

MATERIALS AND METHODS

A blood sample was obtained during standard genetic counseling at the Laboratory of Genetic Epidemiology of the Research Centre for Medical Genetics, Russian Academy of Medical Science, Moscow, Russian Federation. The study was approved by its Internal Review Board (IRB).

DNA was isolated with the salting out procedure [9]. The polymerase chain reaction (PCR) mixtures contained 50 pg of the template, 1 nM

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each of the forward and reverse primers, 2.5 mM MgCl₂, 0.2 mM dNTP mix, 0.5 units of Taq DNA polymerase, and 1 ' Taq buffer in a total volume of 10 ml. All samples were denatured for 1 min. at 94°C, followed by 35 cycles of denaturation at 94°C for 10 seconds, annealing at 62°C for 5 seconds, and extension at 72°C for 90 seconds in a Tercyc thermocycler (DNA-Technology, Moscow, Russia). Oligonucleotides were synthesized by Lytech Inc., Moscow, Russian Federation (<http://www.lytech.ru>); forward: 5'-TGT AAT ATC ACC GTG TCA CTT-3' and reverse: 5'-CAA ACT CTT CAG GCT ACA GAA-3'. Bidirectional sequencing of PCR fragments was performed according to the fluo-rescence-tagged chain termination protocol (BigDye Terminator; Applied Biosystems, Carlsbad, CA, USA) followed by fractionation on an ABI PRISM™ 310 automated DNA sequencer (Applied Biosystems).

RESULTS AND DISCUSSION

A 28-year-old Russian woman was referred to the Research Centre for Medical Genetics, Moscow, Russian Federation in June 2007, because of alopecia, palmoplantar hyperkeratosis and nail dystrophy (Figure 1). She mentioned that she had the above symptoms from early childhood. Moreover, she noted that her mother and maternal grandfather had the same problem since their birth. During clinical examination it was diagnosed as hypohidrotic ectodermal dysplasia, alopecia (with very sparse hair on her scalp, length about 15-20 cm, no eyelashes, eyebrows, axillary and pubic hair), clubbing of the fingers and palmar hyperkeratosis with dystrophy and hypoplasia of the nails, progressive corneal dystrophy (with photophobia and conjunctivitis). The patient showed normal teeth and intellectual development. Light microscopy of hair on the scalp



Figure 1. Clinical presentation. (a) Generalized hypotrichosis; (b) nail dystrophy on the hand; (c) nail dystrophy on the foot; (d) plantar hyperkeratosis.

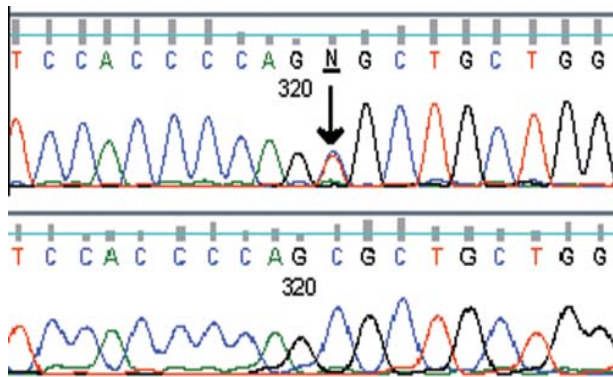


Figure 2. Chromatograms of *GJB6* sequencing of the proband (upper) and a normal child (lower). The arrow indicates the c.263C>T mutation.

revealed thin hair, smooth hair shaft with absence of structural defects in the examined sample. The patient was worried about the risk of having an affected child.

During genetic counseling, Clouston syndrome was hypothesized and the woman was sent for molecular genetic screening of mutations in the *GJB6* gene. We performed whole gene sequencing using primers flanking the open reading frame of the gene, which revealed a heterozygous non synonymous substitution c.263C>T (Figure 2, upper chromatogram). At the protein level, the mutation leads to the amino acid substitution p.A88V. This is one of the mutations found in patients with HED2.

In-depth analysis of the pedigree of the proband's family revealed that at least one person in each generation was affected (Figure 3). This information supported the autosomal dominant type of disease inheritance. Unfortunately, all other affected relatives were dead so we were unable to perform medical and molecular genetic analyses on them. Nevertheless, the patient decided to have a child. The 32-year-old husband of the proband was normal. Thus, the risk of the birth of an affected child was 50% in this case. The pregnancy was unremarkable. During prenatal screening of chorionic villi, the child was determined to have a normal genotype of the *GJB6* gene (Figure 2, lower chromatogram). The child's parameters at birth were within the normal range.

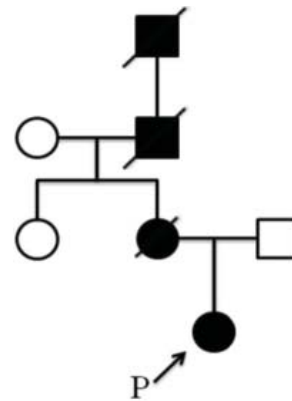


Figure 3. Pedigree of the proband's family. Filled symbols indicate affected individuals.

CONCLUSIONS

This very interesting case of Clouston syndrome in Russia raised more questions than answers. First of all, we genotyped the patient. The genotyping revealed a heterozygous c.263C>T mutation (p.A88V). Neither this mutation nor Clouston syndrome has been described in Russia before.

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