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Variability of auditory threshold at deaf patients with splice site c.-23+1G>A mutation in GJB2 gene (Konneksin 26)

ABSTRACT

In this article results of the audiological examination testifying to auditory threshold variability at deaf patients with splice site c.-23+1G>A mutation in *GJB2* gene in homozygous state are presented. According to this study this *GJB2*-genotype is characterized by (horizontal) flat orsloping audioprofile. There are recommendations for applying the results obtained in practice.

Keywords: deafness, c.-23+1G>A, *GJB2* gene, audiometric analyses, hearing thresholds.

INTRODUCTION

According to preliminary data of the audiological screening in the Republic of Sakha (Yakutia), the frequency of congenital hearing loss is higher than world average value (1: 1000 of newborns) [8, 9] and it is approximating to 1 per 700-900 infants [3]. At the same time, the prevalence of autosomal recessive deafness caused by homozygous c.-23+1G>A mutation of the *GJB2* gene is estimated as 16.2 per 100,000, and the carrier frequency of this mutation is one of the highest in the world (3% - 11% of the indigenous population of Yakutia are heterozygous c.-23+1G>A carriers). Thus, the territory of Eastern Siberia is known as the territory of extensive accumulation of c.-23+1G>A mutation in *GJB2* gene [3].

Several studies of *GJB2* gene mutations in Europe and Asia show a large variability of hearing thresholds in individuals with different mutations in the *GJB2* gene [2, 4, 7, 10, 11, 13]. However, in present time studies of hearing status in individuals homozygous for the c.-23+1G>A mutation in *GJB2* gene are rare. From available data of audiological analysis of three siblings from Bangladesh with c.-23+1G>A mutation in homozygous state are known, that two siblings had a moderate hearing loss, one – profound hearing loss, and audiological curve had a flat and sloping form [2]. Also, we had previously described audiological characteristics of 40

homozygous patients with splice site c.-23+1G>A mutation in *GJB2* gene from Yakut population [3].

Data on hearing thresholds in extended sample of deaf individuals with homozygous c.-23+1G>A mutation in *GJB2* gene are presented in this study. Results of this study will be useful for audiological and rehabilitating aid.

MATERIALS AND METHODS

Patients

108 deaf Yakut individuals (216 ears, the average age of 14.32±4.7 years) with a confirmed genotype c.-23+1G>A/c.-23+1G>A were included in studied sample.

Mutation analysis of GJB2 gene

A total 108 samples of genomic DNA, extracted from leukocytes of peripheral blood, were used for *GJB2* gene mutation analysis. Amplification of the coding (exon 2) and noncoding (exon 1) and flanking intronic regions of *GJB2* gene was conducted by PCR on thermocycler «MJ Mini» (Bio-Rad) using appropriate primers [5,6-15]. The PCR products were subjected to direct sequencing using the same primers on ABI PRISM 3130XL (Applied Biosystems, USA) («Genomics» Core Facility; Institute of Chemical Biology and Fundamental Medicine, Siberian Branch of the Russian Academy of Sciences, Novosibirsk, Russia).

Audiological examination

Audiological examination was conducted on all 108 individuals (216 ears). Air conduction thresholds were measured on frequencies 0.25, 0.5, 1.0, 2.0, 4.0, 8.0 kHz and bone conduction thresholds – on frequencies 0.25, 0.5, 1.0, 2.0, 4.0 kHz with audiometer «MAICO ST20» (MAICO, Germany) in the same conditions for all participants. The results were obtained separately for each ear on air conduction thresholds as on all measured frequencies, and also on speech range of frequencies – PTA 0.5,1.0,2.0,4.0 kHz separately.

Ethical approval

Written informed consent was obtained from all individuals. This study was approved by the local Committee on Biomedical Ethics of Yakut Scientific Center of Complex Medical Problems, Yakutsk, Russia (Yakutsk, Protocol No 16, April 16, 2009).

RESULTS AND DISCUSSION

Data from hearing thresholds audiograms of all studied patients (108 individuals, 216 ears) were summarized and presented in Figure 1.

The upper limit (minimum audiometric values) of hearing thresholds in the range of «low» frequencies (0.25 - 0.5 kHz) was estimated at 20.0 dB, «medium» frequency range (1.0 - 2.0 kHz) - 35.0 dB, «high» frequencies (4.0 - 8.0 kHz) - 40.0 dB. The lower limit (maximum audiometric values) was limited to 105.0 - 120.0 dB (Figure 1).

In the majority of studied ears (83.79%) hearing sensitivity was preserved at all measured frequencies (up to 8.0 kHz inclusive) and only 16.21% of the cases had a loss: 16 ears (7.41%) - 4.0 kHz, 6 ears (2.78 %) - 2.0 kHz and 13 ears (6.02%) - 1.0 kHz (Figure 1).

Hearing thresholds percentiles were examined to determine the audioprofile characteristic for presence of mutation 23+1G>A. The 25th, 50th and 75th percentiles were calculated. The audiological curves at the 25th and 75th percentile were presented by sloping curves, while the 50th percentile (mean) was close to the flat curve, and a wide interval (from 35.0 dB to 45.0 dB at different frequencies) between the 25th and 75th percentiles (Figure 1) confirmed the variability of hearing loss.

The results of audiological analysis of three individuals with a homozygous splice site c.-23+1G>A mutation in the *GJB2* gene from Bangladesh [2], are comparable with our results. Preservation of the remaining hearing of the majority of studied individuals with genotype c.-23 +1G>A/c.-23+1G>A, is consistent with the results of a multicenter study [13], where compound-heterozygous c.35delG/c.-23+1G>A individuals presented significantly less severe hearing loss than homozygous c.35delG/c.35delG patients [13].

Clinical recommendations

The flat audioprofile in individuals with *GJB2*-genotype c.-23+1G>A/c.23+1G>A should be considered in logopaedic and speech training programs. For selecting and programming of hearing aid for children more effective habilitation and rehabilitation is possible considering results of this study.

CONCLUSION

The results of this study demonstrate variability of hearing thresholds in individuals with homozygous c.-23+1G>A mutation in the *GJB2* gene. In addition, auditory sensitivity in the measured frequencies (0.25, 0.5, 1.0, 2.0, 4.0, 8.0 kHz) was remaining intact. Calculation of the 25th, 50th and 75th percentiles showed that presence of mutation c.-23+1G>A is reflected in flat or sloping audioprofile.

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