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ASSESSMENT OF HIGH FREQUENCY 35DELG MUTATION IN GJB2-RELATED DEAFNESS SYNDROME IN A POPULATION WITHOUT TABRIZ CITY, IRAN

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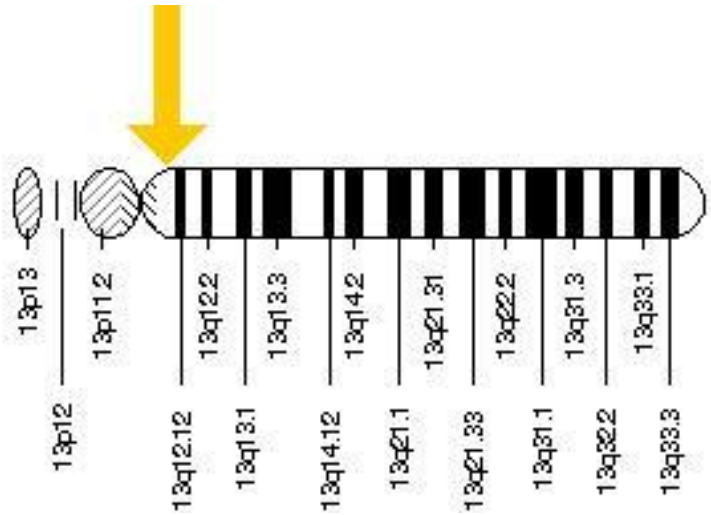
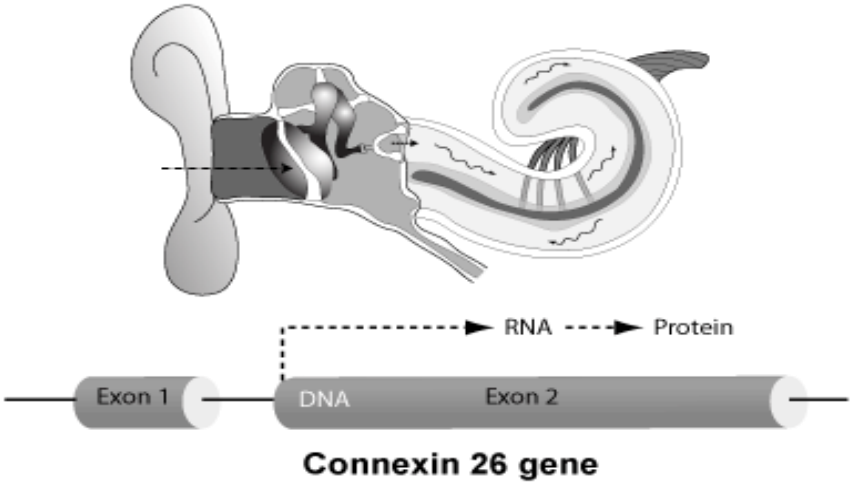
ABSTRACT
Research conducted in different parts of the country reflects the importance of the 35delG mutation in the GJB2 gene. But the mutations listed in the incidence of non-syndromic deafness with autosomal recessive inheritance pattern (ARNSHL) in the city of Tabriz is unknown. In this study, the mutation frequency in the population are non-syndromic hearing was conducted in Tabriz. Non-syndromic recessive deafness Tabrizi 79 non-related patients were studied. Screening of GJB2 35delG mutation that causes some cases of this type of hearing loss is considered, using molecular techniques ARMS - PCR (Allel Refraction Mutation System - Polymerase Chain Reaction) and the nucleotide sequences were determined. 137 studied chromosomes 35delG mutation was detected in 43 chromosomes. 35delG mutation in most areas of the country, especially in the North and North West, high frequency and thus to be based in the North West of the city of Tabriz, Tabriz, the mutation frequency in the population is too common.

KEY WORDS: deafness, mutation 35delG, autosomal recessive, protein CX26, Tabriz.

INTRODUCTION
birth prevalence of congenital deafness heterogeneous disorder that is 1 in 1000 births[1]. Because half the genetic and acquired factors make up the other half. Hearing impairment may be associated with other symptoms appear or only symptom of the disease[2]. Almost 70% of genetic deafness, autosomal recessive inheritance pattern without the syndrome (85%), autosomal dominant (12-15%) or related to X (% 1-3) to[3]. Many genes involved in hearing loss, most notably the GJB2 gene protein connexin 26 (CX26) is encrypted. This gene is located on the long arm of chromosome 13 and exon 1 and intron 2 is formed.



Exon 2, only the GJB2 gene codon connexin 26 is necessary for protein synthesis. Protein connexin 26 channels open connections β2 is a member of the family. The connection structure of the cell membrane, adjacent cells in multicellular organisms are created and allows molecules smaller than 1KD cells pass through[4].



Connexin 26 gene

CX26 protein is present in the human cochlea in the inner ear. GJB2 mutations at positions 30 and 35, there are six replicates guanine nucleotide deletion of nucleotides in this region are the most common causes or 30delG mutation is called 35delG. The major cause of sporadic and hereditary congenital deafness in the white population, the frequency of the 35delG mutation carriers heterozygous for this mutation in northern Europe% 1/ 26 and in southern Europe %1/96 report[5].

Mutation (SNP)	Protein	Gene	References
V37I	Connexin26	GJB2	Kelley et al. (12), Snoeckx et al. (13)
R143W	Connexin26	GJB2	Brobby et al. (14)
235delC	Connexin26	GJB2	Park et al. (11), Dai et al. (15)
A1555G	12S rRNA	MTRNR1	Bae et al. (8)
IVS7-2A>G	Pendrin	SLC26A4	Coucke et al. (16), Park et al. (6)
IVS9+3A>G	Pendrin	SLC26A4	Park et al. (5)
H723R	Pendrin	SLC26A4	Usami et al. (17), Park et al. (6)

MATERIALS AND METHODS

In this study, 79 of the deaf and hard of hearing were genetically determined. The subjects were Rondom intercourse. The clinical condition of the ear, nose and throat was confirmed. Selection factors are deaf or hearing impaired individuals were evaluated with non-syndromic autosomal recessive inheritance pattern, which was approved by the audio metric method was performed. After obtaining informed consent from the patient, the amount of 5-10 ml of venous blood was taken for DNA extraction from them. Samples in order to prevent clotting, were kept in tubes containing EDTA. Genomic DNA was extracted by standard methods Miller. 35delG mutation detection techniques ARMS-PCR using specific primers and standard methods were used. In positive cases, the nucleotide sequence of exon 2 mutations in the GJB2 gene was confirmed by ABI737 system.

Table 1: nucleotide sequences of the genes involved in deafness syndrome disorder ARMS-PCR.

Nucleotide sequence	Gene name
5'TTGGGGCACGCTGCAGACGATCCTGGGGAG3'	**35delGN**
5'TTGGGGCACGCTGCAGACGATCCTGGGGAT3'	**35delGM**
5'GAAGTAGTGATCGTAGCACACGTTCTTGCA3'	**35delGR**
5'GGGCCTCAGTCCCAACATGGCTAAGGTG3'	**CAAT3**
5'CCCACCTTTTCCCCTCTCCAGGCAAATGGG3'	**CAAT4**

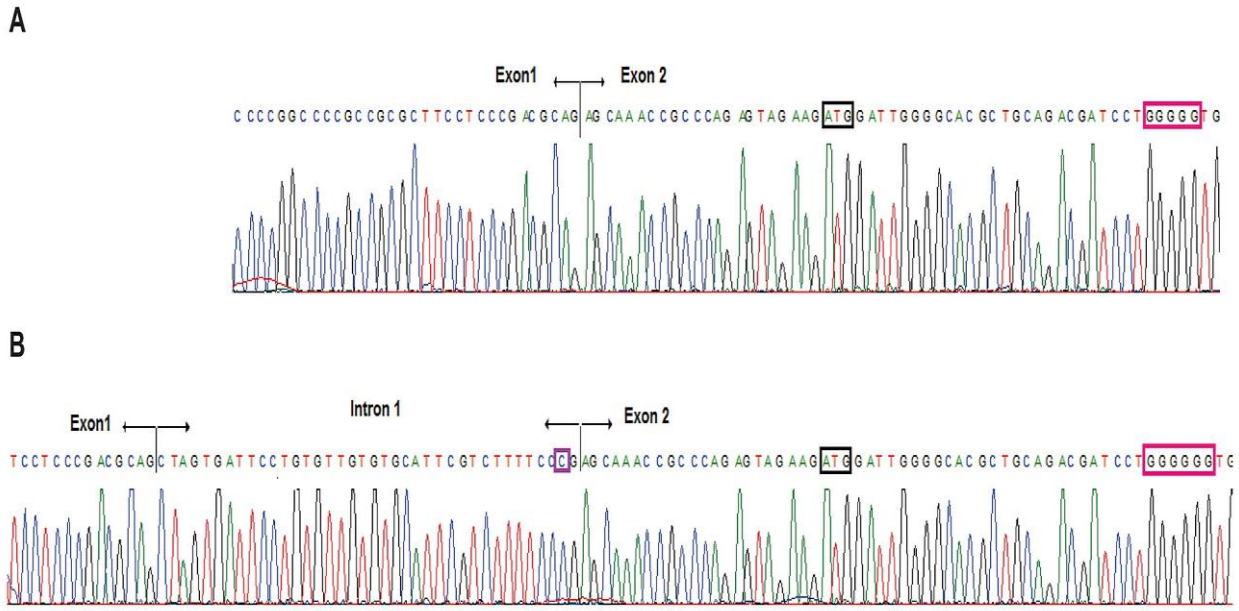


Figure 1: Diagram of the nucleotide sequence of the gene; GJB2 sequence in exon 1 and 2.

Findings

137 chromosomes from 79 patients were examined. The 35delG mutation was found in 47 chromosomes. The chromosomes of the 31 patients, 16 of the homozygote (32 chromosomes) and 15 heterozygous individual (chromosome 15), respectively.

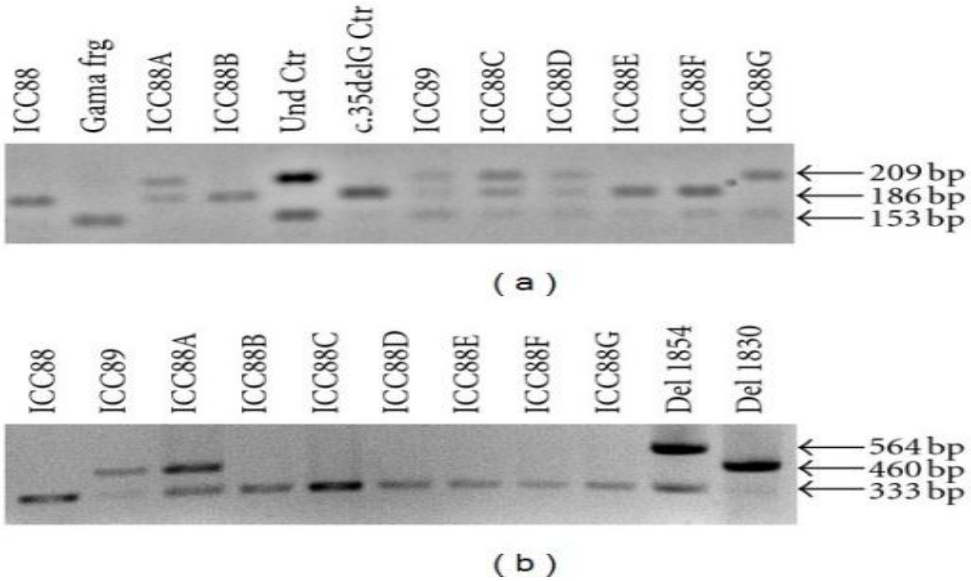


Figure 3: The pattern formed in the band of target genes for congenital deafness.

DISCUSSION AND CONCLUSION

GJB2 gene mutations in more parts of the world, causing mild to severe hearing loss is 50%. Specific mutations of this gene are common in different populations.[6] For example, mutations in Ashkenazi Jews 167delT, in East Asia mutation 235delC, and in many European countries, the prevalence of 35delG mutation[7]. The 35delG mutation in populations of the world's most frequently seen differently. Iran is composed of many nations, according to the frequency of GJB2 mutations in different nations, It is therefore important that different ethnicities and races vary country to be evaluated separately[8]. The basic research on the Iranian population, the frequency of mutation 35delG from 0% to of Sistan and Baluchestan% 27/1 in Gilan been different. The allele frequency of this mutation in the project,% 28/6 in the whole country which is a lot. Therefore, given the frequency of this mutation, mutations in the GJB2 gene or other genes involved assessment in this type of deafness in most areas of the country, the genetic profile of this hearing is completed. As a result of using the information, genetic counseling before pregnancy to family needs.

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