Non-syndromic recessive hearing loss Linkaged TMPRSS3 gene in the Turkish population

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Özet

Türk Populasyonundaki TMPRSS3 Genine Bağlı Non-Sendromik Resesif İşitme Kaybı

Canlı doğumlarda yaklaşık 1/1000 sıklıkla görülen konjenital sağırlıkların %50'sinin temelinde genetik nedenler yatmaktadır. Non-sendromik işitme kayıpları kalıtım kalıpları baz alınarak sınıflandırıldıklarında, vakaların %80'inin otozomal resesif kalıtım sergilediği gözlenmektedir. Bu çalışmada non-sendromik otozomal resesif, prelingual işitme kaybına sahip ve yüksek oranda (%77) akraba evliliğinin görüldüğü 26 Türk ailede TMPRSS3 geni ile bağlantının varlığı araştırıldı. Bağlantı ve lod-score analizlerinden elde edilen verilerle 26 aileden altısında (%23) TMPRSS3 genine potansiyel bağlantı saptandı. TMPRSS3 geninin Türk populasyonundaki non-sendromik otozomal resesif sağırlığın başlıca nedenlerinden olabileceği gösterildi. Bu çalışma ile Türk ailelerde TMPRSS3 genine bağlantı ilk kez rapor edilmektedir.

Anahtar kelimeler: Resesif işitme kaybı, TMPRSS3 geni

Abstract

Congenital deafness occurs in approximately 1 in 1000 live births and it is estimated that at least 50% of these cases are due to genetic cause. Non-syndromic deafness is classified according to its mode of inheritance and non-syndromic autosomal recessive deafness accounts for approximately 80% of congenital hereditary deafness case. In this study we present evidence for linkage to TMPRSS3 gene in 26 non-syndromic autosomal recessive prelingual hearing impaired Turkish families with high frequency of consanguinity. Linkage and lod-score data suggesting that 6 of 26 (23%) Turkish families showed potential linkage to TMPRSS3 gene. The findings imply that TMPRSS3 might be one of the major contributory genes to non-syndromic recessive prelingual deafness in Turkish population. Linkage to TMPRSS3 gene in Turkish patients is reported for the first time in this study.Recessive hearing loss, TMPRSS3 gene

Anahtar kelimeler: Recessive hearing loss, TMPRSS3 gene

Introduction

Hearing loss can lead to alterations in language, speech, cognitive and psychosocial development, and it is the most common sensory defect of humans with an incidence of about 1 child in 1000 born (1). Hearing loss can be classified as genetic or non-genetic, prelingual or postlingual, and syndromic or non-syndromic. It can be genetic, resulting from a mutation in a single gene (monogenic forms) or from a combination of mutations in different genes and environmental factors (multifactorial forms). Approximately 50% of cases are due to monogenic forms of hearing loss (1-3). The inheritance pattern of monogenic prelingual non-syndromic hearing loss is autosomal recessive in approximately 80% of patients (4, 5).

Recently, linkage analyses have mapped many genes for non-syndromic hearing loss to different chromosomes. Over 30 loci are known to be responsible for autosomal recessive inheritance of the non-syndromic hearing impairment. Two of the previously reported loci for non-syndromic recessive deafness are DFNB8 and DFNB10, both located on chromosome 21q22.3 (6). TMPRSS3 or ECHOS1 gene was found to be responsible for both the DFNB8/DFNB10 phenotypes with congenital/childhood non-syndromic hearing loss (7-9). TMPRSS3 encodes a transmembrane serine protease, and its expression was demonstrated in fetal cochlea and many other tissues. TMPRSS3 may be involved in the development and maintenance of the inner ear (9, 10). In this study we have studied 26

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non-syndromic autosomal recessive prelingual hearing impaired Turkish families with high frequency of consanguinity.

Materials Methods

Materials: To determine the prevalence of TMPRSS3 deafness in Turkish patients, a total of 26 families with severe to profound sensorineural prelingual non-syndromic deafness and with high frequency of consanguinity (77%) suggested an autosomal recessive mode of inheritance with full penetrance at birth was included. There was no evidence for an autosomal dominant or X-linked mode of inheritance or any obvious syndrome. The families that segregate congenital deafness are either large enough to support statistically significant linkage or have at least two affected subjects. Families were ascertained in schools for the deaf in northeast Turkey. Informed consents was obtained for all participating family members.
Methods: Peripheral blood was obtained from members of all 26 families with their permision, and genomic DNA was isolated from blood lymphocytes by the standard protocol (11). Marker information used for DFNB8/B10 region was provided by the Hereditary Hearing Loss Homepage (http://www.uia.ac.be/dnalab/hhh). Two highly polymorphic markers generated by Généthon were used. D21S212 and D21S1225 are tightly link to TMPRSS3 gene (D21S1225 -0.1 cM- TMPRSS3/ D21S212). Amplification reaction (12,5 µl) was performed with 100 ng genomic DNA and 2,0 pmol each primer. Cycling parameters consisted of 94ºC 5 min, followed by 36 cycles of 94ºC (35 sec), 55ºC (35 sec), 72ºC (35 sec) and finally 72ºC 2 min. Amplification products were run on a 6% (19:1, acrilyamide:bisacrylamide) denaturating polyacrilamide gel. Samples were electrophoresed for 4 hours at 1200 Volt at room temperature then silver staining was performed.
Haplotypes were constructed for all persons in the 26 pedigrees by using the chromosome 21 markers adjacent to TMPRSS3 gene on 21q22.3, D21S212 and D21S1225, included in the multilocus analyses. Using the computer program MLINK two-point linkage analyses were permormed. The disease was modeled as an autosomal recessive trait with complete penetrance at birth. Complications of two-point LOD-Scores (Z) in these families were facilitated by reducing the number of alleles for each marker system to no more than ten. Inbreeding loops were preserved in the pedigrees.

Results

We have ascertained 26 families in Turkey consisting of 177 members, of which 69 were affected. Affected and unaffected members of each family were clinically evaluated for the presence or absence of deafness and pure tone audiometry. The results and the defect were compatible with congenital, recessive, severe (70<ACPTA90 dB) to profound (ACPTA >90 dB) sensorineural deafness. Deafness was congenital and no additional phenotypes such as heart problems, morphological defects in outer ear, defects in other organs, or mental retardation were observed. Audiometric results of unaffected sibs were normal. The familial pattern of congenital hearing loss and high frequency of consanguinity in each family suggested an autosomal recessive mode of inheritance with full penetrance at birth. 20 of 26 (77%) Turkish families contained several consanguineous marriages. The segregation of D21S212 and D21S1225 polymorphic microsatellite markers were tested for linkage to TMPRSS3 gene in 26 families. We have constructed haplotypes of all family members using the alleles at loci D21S212 and D21S1225. No recombinants are evident for D21S212 and D21S1225. Linkage to TMPRSS3 gene was detected in six of twenty-six (23%) Turkish families, so the other 20 families were excluded. Also amplification products of these six potentially linked family members, were run on the same gel and seen that they don't show the same alleles. So we concluded that these six families don't have a common haplotype and are not consanguineous. The markers showed significant evidence for linkage in families (Table). The pedigrees of the families are shown on Figure1-6.

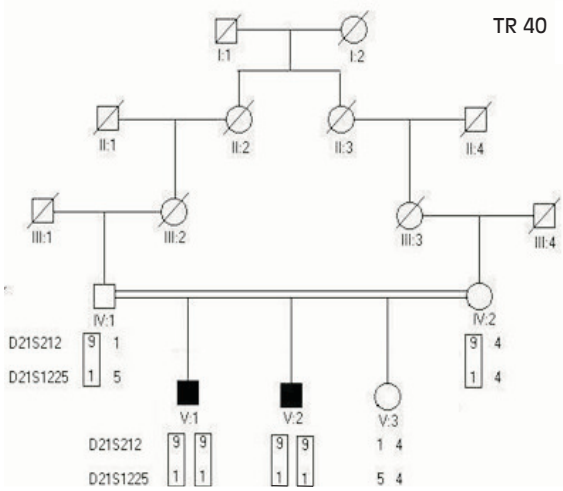
Discussion

Approximately 50% of recessive non-syndromic deafness can be attributed to mutations in the connexin26 (GJB2) gene in many Caucasians populations. However, the prevalence of GJB2 gene mutations among deaf in Turkey is low (12, 13), raising the possibility that other genes may contribute to prelingual non-syndromic recessive deafness in Turkish population.
Previously reported TMPRSS3 mutations have been identified in a Palestinian, five Pakistani, and two Tunisian families and in Caucasian patients from Spain, Italy, Greece, and Australia (7-10). The findings of this study imply that TMPRSS3 might be one of the major contributors to the prelingual hearing loss

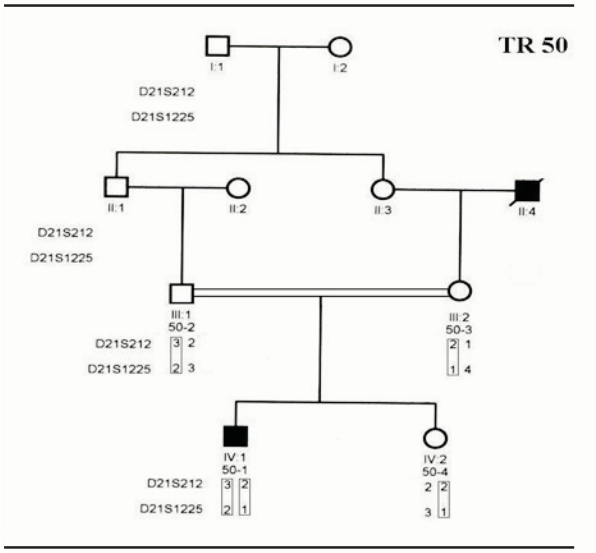
Table 1: Results of pair wise linkage analyses between TMPRSS3 and two markers (D21S212 and D21S1225) in seven potantially linked families.

Family	Marker	Z at θ					
		0.00	0.05	0.1	0.2	0.3	0.4
40	D21S212	0.727	0.639	0.549	0.367	0.193	0.055
	D21S1225	0.727	0.639	0.549	0.367	0.193	0.055
50	D21S212	0.125	0.086	0.056	0.018	0.004	0.000
	D21S1225	0.125	0.086	0.056	0.018	0.004	0.000
53	D21S212	1.646	1.439	1.228	0.803	0.409	0.112
	D21S1225	0.857	0.744	0.630	0.408	0.208	0.057
69	D21S212	0.301	0.258	0.215	0.134	0.064	0.017
	D21S1225	0.301	0.258	0.215	0.134	0.064	0.017
70	D21S212	0.301	0.258	0.215	0.134	0.064	0.017
	D21S1225	0.602	0.515	0.430	0.267	0.129	0.034
88	D21S212	1.492	1.298	1.099	0.696	0.329	0.080
	D21S1225	1.492	1.298	1.101	0.705	0.344	0.088

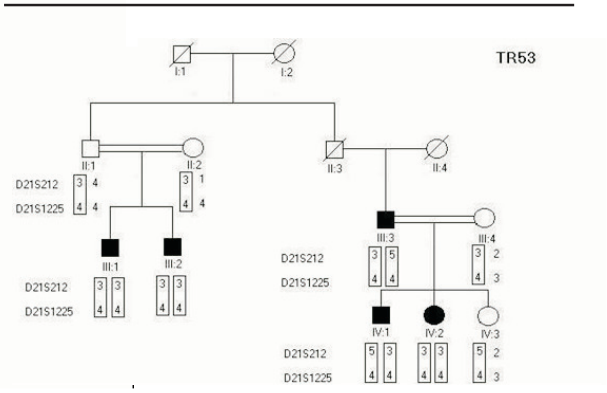
in the Turkish population with prevalence of 27%. However, TMPRSS3 is still an important contributor to genetic deafness in Turks. This compares to 5 of 160 (3%) of Pakistani families studied with TMPRSS3 mutations and large isolated Palestinian and Tunisian families described before (10). In 448 unrelated deaf patients from Spain, Italy, Greece and Australia who did not have GJB2 mutations it has been shown that TMPRSS3 mutations contribute to less than 1% of nonsyndromic childhood deafness in Caucasians (10). In a similar panel of 64 deaf North American individuals no TMPRSS3 mutations were detected (9). Thus TMPRSS3 gene is potentially "one of the most frequent Turkish deafness genes" albeit rare in other populations.



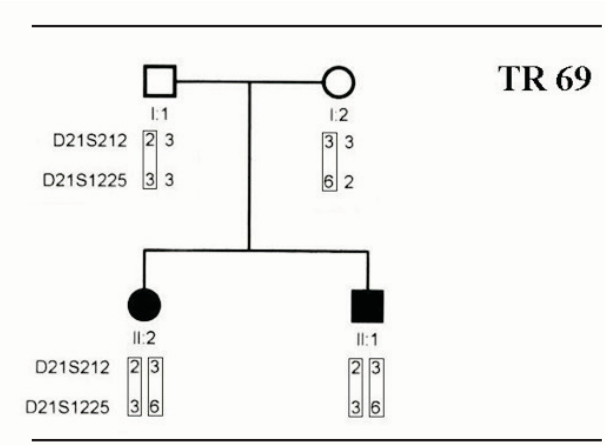
Figüre 1



Figüre 2



Figüre 3



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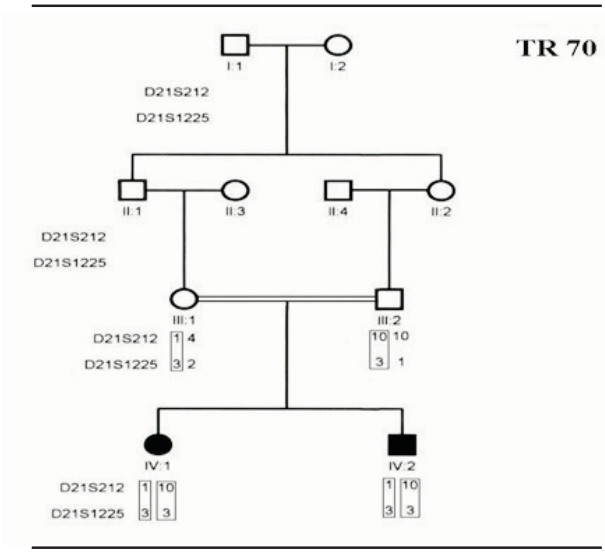
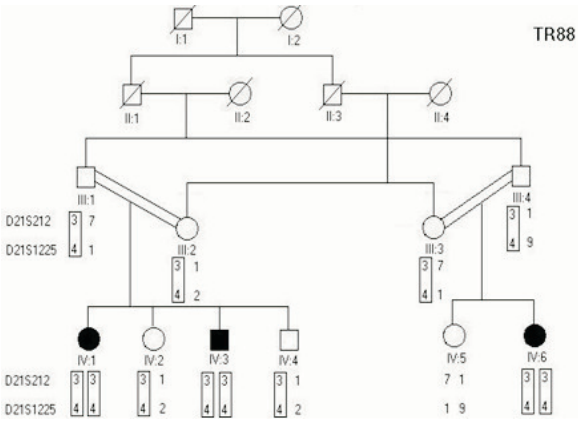


Figüre 5



Figüre 6

Figures 1-6: Pedigrees of hearing impaired Turkish families (TR40, TR50, TR53, TR69, TR70 and TR88) showed potential linkage to TMPRSS3 gene and genotype data for markers D21S212 and D21S1225. Boxes identify specific alleles.

According to recent data about TMPRSS3 gene prevalence in other populations, it is high prevalence of its in the Turkish population is remarkable. The results of this study can be supported by other researches if should be done countries in Middle East, Middle and Far Asia. Another result of the study is an evident for migration from central Asia to Europe, especially Anatolia. The findings may be importan implication for genetic counseling and also human migration events, both genetic and history science. Hearing disorders represent a significant health problem in the world population. Thus, identification of the gene defect causing deafness and the understanding of the gene products will contribute significantly to the understanding of the molecular events that are important for hearing function (4). In our study further work is needed to screen the gene in families showing potential linkage to TMPRSS3 and identify TMPRSS3 substrates and the role it plays in hearing process.

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