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DFNB89, a novel autosomal recessive nonsyndromic hearing impairment locus on chromosome 16q21-q23.2

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Abstract

DFNB89 is a novel autosomal recessive non-syndromic hearing impairment (ARNSHI) locus that was mapped to 16q21-q23.2. Linkage to the region was established by carrying out genome-wide linkage scans in two unrelated, consanguineous Pakistani families segregating ARNSHI. The maximum multipoint LOD score is 9.7 for both families and for each family, a significant maximum LOD score of 6.0 and 3.7 were obtained. The 3-unit support interval and the region of homozygosity for the two families extend from rs717293 (chr16: 62.1 Mb) to rs728929 (chr16: 78.2 Mb) and contain 16.1 Mb of sequence. A total of 146 genes are within the DFNB89 interval. Eight candidate genes, *CALB2*, *CDH1*, *CDH3*, *CDH11*, *HAS3*, *NOB1*, *PLEKHG4* and *SMPD3*, were sequenced, but no potentially causal variants were discovered. DFNB89 is the second ARNSHI locus mapped to chromosome 16.

Introduction

Currently, ~150 loci for nonsyndromic (NS) hearing impairment (HI) have been mapped, and >50 NSHI genes have been identified, of which 37 genes are involved in the genetic etiology of autosomal recessive (AR) NSHI (Hereditary Hearing Loss Homepage). The discovery of NSHI genes and their causal variants aid in both diagnostic genetic screening and therapeutic interventions for HI as well as increase our understanding of the pathophysiology of HI.

DFNB89, a new locus for ARNSHI, was mapped to chromosome 16q21-q23.2 through a genome-wide linkage scan and homozygosity mapping using 21 DNA samples from two unrelated, consanguineous Pakistani families. Each family could independently establish linkage to the region with significant maximum multipoint LOD scores of 6.0 and 3.7. Within the DFNB89 region, each family has a unique haplotype, and the overlapping regions of homozygosity resulted in an interval containing 16.1 Mb of sequence.

Materials and methods

Family history and clinical evaluation

With the prior approval from the Institutional Review Boards of the Quaid-I-Azam University and the Baylor College of Medicine and Affiliated Hospitals, the study was initiated and informed consent was obtained from all family members who participated in the study. Families 4338 (Fig. 1) and 4406 (Fig. 2) are consanguineous kindreds displaying ARNSHI. These families are from Azad Jammu and Kashmir in the Pakistani-administrated north-western region of the Indian subcontinent.

The ARNSHI in both families is bilateral, non-progressive, prelingual, and moderate-to-severe affecting all frequencies. Perinatal and maternal history and interviews regarding infections, ototoxic medication, head trauma and noise exposure did not indicate a possible environmental cause for the HI. Following careful evaluation for systemic features, no obvious stigmata that would indicate that the HI is syndromic were found. Romberg testing and gait assessment were both negative. Fundoscopic examination did not reveal any evidence of retinitis pigmentosa.

DNA extraction, genotyping and linkage analysis

For family 4338, venous blood was obtained from 12 family members, 7 of whom are hearing impaired (Fig. 1), while for family 4406, 4 of 9 pedigree members who provided a blood sample present with HI (Fig. 2). Genomic DNA was extracted following a standard protocol (Grimberg et al. 1989). A whole genome linkage scan was carried out at the Center for Inherited Disease Research (CIDR) using the Illumina Human Linkage-12 panel which contains 6,090 SNP marker loci.

On the generated genotype data, PedCheck (O'Connell and Weeks 1998) was used to identify Mendelian inconsistencies, while the MERLIN (Abecasis et al. 2002) program was used to assess the data for occurrence of double recombination events over short genetic distances, which are most likely due to genotyping error. Two-point linkage analysis was carried out using MLINK of the FASTLINK package (Cottingham et al. 1993) while multipoint linkage analysis was performed using Allegro1.2c (Gudbjartsson et al. 2000). An AR mode of inheritance with complete penetrance and a disease allele frequency of 0.001 were used in the analysis. Marker allele frequencies were estimated from observed and reconstructed genotypes of the founders from families 4338 and 4406 and 49 additional Pakistani families that under-went a genome scan at CIDR. For multipoint linkage analysis, genetic map positions according to the Rutgers combined linkage-physical map of the human genome (Build 36 version) (Matisse et al. 2007) were used. The March 2006 human reference sequence (Build 36) was used as the reference map to determine the physical positions of SNP marker loci, which were not available on the Rutgers map, and then interpolation was used to determine the genetic map distances between the SNP marker loci. Haplotypes were reconstructed using Simwalk2 (Sobel and Lange 1996).

Candidate gene sequencing

Using Primer3 software (Rozen and Skaletsky 2000), primers were designed to sequence the exons, intron boundaries and promoter regions for eight candidate genes: *CALB2*, *CDH1*, *CDH3*, *CDH11*, *HAS3*, *NOB1*, *PLEKHG4* and *SMPD3*. From each family, DNA samples from one hearing and two hearing-impaired family members underwent sequencing [pedigree 4338: IV-3, V-4 and V-6 (Fig. 1) and pedigree 4406: III-1, IV-2 and IV-4 (Fig. 2)]. PCR-amplified DNA products were purified with ExoSAPIT (USB Corp., Cleveland, OH, USA). Sequencing was performed with the BigDye Terminator v3.1 Cycle Sequencing Kit and Applied Biosystems 3730 DNA Analyzer (Applied Biosystems Inc, Foster City, CA, USA). The DNA sequences were assembled and analyzed using the Sequencher software V4.9 (Gene Codes Corp., Ann Arbor, MI, USA).

Results

No genotyping errors were detected through the occurrence of Mendelian inconsistencies or double recombination events occurring over short genetic map distances. A maximum two-point LOD score of 4.5 ($\theta = 0$) for family 4338 was obtained at the marker locus rs1018910 (69.8 Mb), while for family 4406, a maximum two-point LOD score of 2.8 ($\theta = 0$) was observed at rs235987 (69.8 Mb) (Table 1).

To facilitate multipoint linkage analysis, pedigree 4338 was split into two branches, due to its large size. A maximum multipoint LOD score of 6.0 was obtained for nine markers flanked by rs717293 (62.1 Mb) and rs728929 (78.2 Mb) (Table 1). The 3-unit support interval (SI) and the region of homozygosity which spans 20.2 cM is bounded by markers rs717293 (62.1 Mb) and rs728929 (78.2 Mb) and contains 16.1 Mb of sequence (Fig. 1; Table 1). The centromeric boundary of the region of homozygosity is delimited by a recombination event between the marker loci rs717293 and rs973200 in unaffected child V-8 and the telomeric boundary was delimited by a historic recombination event between rs424074 and rs728929 observed in affected children V-1, V-3 and V-4 (Fig. 1).

For family 4406, a maximum multipoint LOD score of 3.7 was obtained for nine markers flanked by rs1482258 (58.1 Mb) and rs1125733 (79.0 Mb). The 3-unit SI and region of homozygosity is flanked by markers rs1482258 (58.1 Mb) and rs1125733 (79.0 Mb) which is 25.4 cM long and contains 20.9 Mb of sequence (Fig. 2; Table 1). The centromeric boundary was delimited by the recombination event between the markers rs1482258 (58.1 Mb) and rs1027277 (61.2 Mb) in the unaffected child IV-5. The telomeric boundary of the

homozygosity region is delimited by a historic recombination event between marker loci rs1079635 and rs1125733 which can be observed in the HI children IV-4 and IV-6 (Fig. 2). The 3-unit SI for family 4338 is completely contained within the 3-unit SI for family 4406 (Table 1).

The 3-unit SI and region of homozygosity are exactly same for family 4338 and contains 16.1 Mb of sequence. DFNB89 was assigned to this locus by the Human Genome Organization (HUGO) Gene Nomenclature Committee.

Within the DFNB89 region, there are 146 known genes including hypothetical proteins. Among those genes within the DFNB89 interval, the protein coding sequences and promoter regions in 5'-UTR of eight candidate genes *CALB2*, *CDH1*, *CDH3*, *CDH11*, *HAS3*, *NOB1*, *PLEKHG4* and *SMPD3* were sequenced. Although several known sequence variants were identified, no potential causal variants were observed.

Discussion

On chromosome 16, 2 NSHI loci have previously been identified: 1 AR, DFNB22 (Zwaenepoel et al. 2002) on 16p12.2 and the other an autosomal dominant (AD) locus, DFNA40, also on 16p12. For DFNB22 Otoancorin (*OTOA*; MIM 607038) (Zwaenepoel et al. 2002) has been identified while the gene for DFNA40 is unknown (Hereditary Hearing Loss Homepage). The mu-Crystallin (*CRYM*; MIM 123740) gene on 16p12.2, which does not have a corresponding locus number, was implicated as being responsible for NSHI in two unrelated individuals (Abe et al. 2003). DFNB89 is the first NSHI locus mapped to the q-arm of chromosome 16 and the third NSHI locus identified on chromosome 16 (Fig. 3).

Although no other NSHI loci map to 16q, the OTSC4 locus for otosclerosis (Brownstein et al. 2006) also maps to 16q and overlaps with the DFNB89 interval (Fig. 3). Although the otosclerosis phenotype is distinct from the NSHI phenotype observed in the two families. It cannot be ruled out that different variants within the same gene is responsible for both OTSC4 and DFNB89. There have been many examples where various forms of HI, e.g. syndromic HI and NSHI are caused by different variants within the same gene.

DFNB89 was mapped in two unrelated, consanguineous Pakistani families. These two kindred do not share the same haplotype (Figs. 1, 2). Therefore, the HI in these families can be due to different causal variants in the same gene or two different genes may be responsible for DFNB89. However, even though two different haplotypes are observed, this does not rule out the possibility that the two families are both homozygous for the same causal variant.

Of the 146 genes in the DFNB89 region, *CALB2*, *CDH1*, *CDH3*, *CDH11*, *HAS3*, *NOB1*, *PLEKHG4* and *SMPD3* were chosen for sequencing. The Cadherin superfamily of genes is associated with both Usher syndrome and ARNSHI (i.e. *CDH23* (Bork et al. 2001) and *PCDH15* (Ahmed et al. 2003)) and facilitates cell–cell adhesion and signaling in various tissues. The Cadherin 11 gene (*CDH11*; MIM 600023) was shown to be necessary for otolith assembly in the developing zebra fish embryo (Clendenon et al. 2009) and was also reported to be expressed in the human fetal cochlea (Skvorak et al. 1999). The Cadherin 1 (*CDH1*; MIM 192090) localizes to the apical intercellular junctions of supporting cells within the murine organ of Corti suggesting a role in inter-supporting cell adhesion (Whitlon 1993). In addition, the Cadherin 3 (*CDH3*; MIM 114021) gene within the DFNB89 interval was sequenced.

Several genes were sequenced because of strong cochlear expression in animal model systems. Calbindin 2 or Calretinin (*CALB2*; MIM 114051) is expressed in inner hair cells,

supporting cells and the cochlear ganglion (Deschene et al. 1991) and has been used as a marker for surviving hair cells and spiral ganglion neurons post-deafness or in differentiating inner ear stem cells (Coppens et al. 2001; Martinez-Monedero et al. 2008). The Hyaluronan Synthase 3 (*HAS3*; MIM 602428) was also sequenced due to gene expression within the otic vesicle of mouse embryo, particularly in the vestibulocochlear ganglion (Tien and Spicer 2005). The NIN1/RPN12-binding Protein 1 (*NOB1*; MIM 613586) has specific expression in spiral ganglion cells and is up-regulated in deaf guinea pigs (Han et al. 2009).

Two genes were selected as candidates for DFNB89 because of known associations with diseases that commonly occur with HI. A deletion in the mouse ortholog of sphingomyelin phosphodiesterase 3, neutral membrane (*SMPD3*; MIM 605777) was recently described to cause osteogenesis and dentinogenesis imperfecta, two diseases that are both associated with hearing loss (Aubin et al. 2005). On the other hand, HI was documented in 38% of patients with autosomal dominant cerebellar ataxia due to Puratrophin 1 or Pleckstrin Homology Domain-containing Protein, Family G, Member 4 (*PLEKHG4*; MIM 609526) (Ouyang et al. 2006).

It remains to be seen if the DFNB89 locus contains more than one HI gene, or a single gene of either typical or variable phenotypic effect. The identification of DFNB89 gene will give us new insight into the hearing mechanism as well as into the genetic etiology of HI.

Electronic database information

The following URLs were accessed for data in this article: Hereditary Hearing Loss Homepage, <http://www.hereditaryhearingloss.org/>, Online Mendelian Inheritance in Man (OMIM), <http://www.ncbi.nlm.nih.gov/omim/>, UCSC genome browser, <http://www.genome.ucsc.edu/>.

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References

- Abe S, Katagiri T, Saito-Hisaminato A, Usami S, Inoue Y, Tsunoda T, Nakamura Y. Identification of CRYM as a candidate responsible for nonsyndromic deafness, through cdna microarray analysis of human cochlear and vestibular tissues. *Am J Hum Genet.* 2003; 72:73–82. [PubMed: 12471561]
- Abecasis GR, Cherny SS, Cookson WO, Cardon LR. Merlin—rapid analysis of dense genetic maps using sparse gene flow trees. *Nat Genet.* 2002; 30:97–101. [PubMed: 11731797]
- Ahmed ZM, Riazuddin S, Ahmad J, Bernstein SL, Guo Y, Sabar MF, Sieving P, Griffith AJ, Friedman TB, Belyantseva IA, Wilcox ER. *Pcdh15* is expressed in the neurosensory epithelium of the eye and ear and mutant alleles are responsible for both *ush1f* and *dfnb23*. *Hum Mol Genet.* 2003; 12:3215–3223. [PubMed: 14570705]
- Aubin I, Adams CP, Opsahl S, Septier D, Bishop CE, Auge N, Salvayre R, Negre-Salvayre A, Goldberg M, Guenet JL, Poirier C. A deletion in the gene encoding sphingomyelin phosphodiesterase 3 (*smpd3*) results in osteogenesis and dentinogenesis imperfecta in the mouse. *Nat Genet.* 2005; 37:803–805. [PubMed: 16025116]
- Bork JM, Peters LM, Riazuddin S, Bernstein SL, Ahmed ZM, Ness SL, Polomeno R, Ramesh A, Schloss M, Srisailpathy CR, Wayne S, Bellman S, Desmukh D, Ahmed Z, Khan SN, Kaloustian

- VM, Li XC, Lalwani A, Bitner-Glindzicz M, Nance WE, Liu XZ, Wistow G, Smith RJ, Griffith AJ, Wilcox ER, Friedman TB, Morell RJ. Usher syndrome 1d and nonsyndromic autosomal recessive deafness dfnb12 are caused by allelic mutations of the novel cadherin-like gene *cdh23*. *Am J Hum Genet.* 2001; 68:26–37. [PubMed: 11090341]
- Brownstein Z, Goldfarb A, Levi H, Frydman M, Avraham KB. Chromosomal mapping and phenotypic characterization of hereditary otosclerosis linked to the *otsc4* locus. *Arch Otolaryngol Head Neck Surg.* 2006; 132:416–424. [PubMed: 16618911]
- Clendenon SG, Shah B, Miller CA, Schmeisser G, Walter A, Gattone VH 2nd, Barald KF, Liu Q, Marrs JA. Cadherin-11 controls otolith assembly: evidence for extracellular cadherin activity. *Dev Dyn.* 2009; 238:1909–1922. [PubMed: 19582870]
- Coppens AG, Kiss R, Heizmann CW, Deltenre P, Poncelet L. An original inner ear neuroepithelial degeneration in a deaf Rottweiler puppy. *Hear Res.* 2001; 161:65–71. [PubMed: 11744282]
- Cottingham RW Jr, Idury RM, Schaffer AA. Faster sequential genetic linkage computations. *Am J Hum Genet.* 1993; 53:252–263. [PubMed: 8317490]
- Deschene CJ, Winsky L, Kim HN, Goping G, Vu TD, Wenthold RJ, Jacobowitz DM. Identification and ultrastructural localization of a calretinin-like calcium-binding protein (protein 10) in the guinea pig and rat inner ear. *Brain Res.* 1991; 560:139–148. [PubMed: 1722130]
- Grimberg J, Nawoschik S, Belluscio L, McKee R, Turck A, Eisenberg A. A simple and efficient non-organic procedure for the isolation of genomic DNA from blood. *Nucleic Acids Res.* 1989; 17:8390. [PubMed: 2813076]
- Gudbjartsson DF, Jonasson K, Frigge ML, Kong A. Allegro, a new computer program for multipoint linkage analysis. *Nat Genet.* 2000; 25:12–13. [PubMed: 10802644]
- Han Y, Hong L, Zhong C, Xue T, He Y, Chen J, Chunyu X, Qiao L, Qiu J. The expression of *NOBI* in spiral ganglion cells of guinea pig. *Int J Pediatr Otorhinolaryngol.* 2009; 73:315–319. [PubMed: 19101043]
- Martinez-Monedero R, Yi E, Oshima K, Glowatzki E, Edge AS. Differentiation of inner ear stem cells to functional sensory neurons. *Dev Neurobiol.* 2008; 68:669–684. [PubMed: 18278797]
- Matisse TC, Chen F, Chen W, De La Vega FM, Hansen M, He C, Hyland FC, Kennedy GC, Kong X, Murray SS, Ziegler JS, Stewart WC, Buyske S. A second-generation combined linkage physical map of the human genome. *Genome Res.* 2007; 17:1783–1786. [PubMed: 17989245]
- O'Connell JR, Weeks DE. Pedcheck: a program for identification of genotype incompatibilities in linkage analysis. *Am J Hum Genet.* 1998; 63:259–266. [PubMed: 9634505]
- Ouyang Y, Sakoe K, Shimazaki H, Namekawa M, Ogawa T, Ando Y, Kawakami T, Kaneko J, Hasegawa Y, Yoshizawa K, Amino T, Ishikawa K, Mizusawa H, Nakano I, Takiyama Y. 16qlinked autosomal dominant cerebellar ataxia: a clinical and genetic study. *J Neurol Sci.* 2006; 247:180–186. [PubMed: 16780885]
- Rozen S, Skaletsky H. Primer3 on the www for general users and for biologist programmers. *Methods Mol Biol.* 2000; 132:365–386. [PubMed: 10547847]
- Skvorak AB, Weng Z, Yee AJ, Robertson NG, Morton CC. Human cochlear expressed sequence tags provide insight into cochlear gene expression and identify candidate genes for deafness. *Hum Mol Genet.* 1999; 8:439–452. [PubMed: 9949203]
- Sobel E, Lange K. Descent graphs in pedigree analysis: applications to haplotyping, location scores, and marker-sharing statistics. *Am J Hum Genet.* 1996; 58:1323–1337. [PubMed: 8651310]
- Tien JY, Spicer AP. Three vertebrate hyaluronan synthases are expressed during mouse development in distinct spatial and temporal patterns. *Dev Dyn.* 2005; 233:130–141. [PubMed: 15765504]
- Whitlon DS. E-cadherin in the mature and developing organ of Corti of the mouse. *J Neurocytol.* 1993; 22:1030–1038. [PubMed: 8106878]
- Zwaenepoel I, Mustapha M, Leibovici M, Verpy E, Goodyear R, Liu XZ, Nouaille S, Nance WE, Kanaan M, Avraham KB, Tekaiia F, Loiselet J, Lathrop M, Richardson G, Petit C. Otoancorin, an inner ear protein restricted to the interface between the apical surface of sensory epithelia and their overlying acellular gels, is defective in autosomal recessive deafness dfnb22. *Proc Natl Acad Sci USA.* 2002; 99:6240–6245. [PubMed: 11972037]

Fig. 2.
Pedigree drawing of family 4406; legend described in Fig. 1

Fig. 3.
The genetic interval of DFNB89 and the locations of other NSHI loci and genes on chromosome 16 are indicated

Table 1

Two-point and multipoint LOD scores for DENB89 families 4338 and 4406

Marker names ^a	Physical position ^b	Genetic position ^c	4338 ^d		4406 ^d	
			Multi-point	Two-point ^e	Multi-point	Two-point ^e
rs1420533	51,121,127	65.99	-∞	-∞	-	-
rs1946155	52,191,796	67.57	-∞	-∞	-	-
rs8060118	53,649,642	71.65	-	-	-3.34	-2.26
rs837529	54,043,795	72.77	-∞	1.23	-1.35	1.38
rs735144	55,441,425	75.12	-	-	-4.27	-2.02
rs1982395	56,226,257	76.74	-5.73	2.40	-	-
rs247041	56,435,141	76.74	-	-	-0.71	1.27
rs6993	57,298,868	78.40	-∞	1.39	-0.92	1.29
rs1482258	58,068,592	79.24	-	-	-0.53	1.56
rs1027277	61,242,497	82.70	-∞	1.13	3.68	2.61
rs717293	62,116,021	83.29	1.90	2.71	3.68	2.14
rs973200	63,565,313	84.76	5.97	3.24	-	-
rs1359839	64,252,852	85.34	-	-	3.68	2.67
rs461785	64,366,545	85.34	5.97	3.65	-	-
rs9033	65,739,500	86.57	5.97	2.86	3.68	2.39
rs235987	69,806,847	87.82	-	-	3.68	2.80
rs1018910	69,835,969	88.19	5.97	4.50	-	-
rs719353	71,600,052	89.73	-	-	3.68	2.40
rs328384	72,397,297	89.91	5.97	4.01	-	-
rs7187229	73,301,237	91.94	5.97	3.31	3.68	1.99
rs11149780	73,497,867	92.48	5.97	4.00	-	-
rs1074964	76,727,759	96.44	-	-	3.67	2.22
rs1079635	77,421,116	97.46	-	-	3.67	1.93
rs1079638	77,467,497	97.46	5.97	3.71	-	-
rs424074	77,792,088	101.04	5.86	3.91	-	-
rs728929	78,227,825	103.48	0.02	-0.46	-	-
rs1125733	79,009,741	104.61	-3.74	-0.72	-4.23	-2.19

Marker names ^a	Physical position ^b	Genetic position ^c	4338 ^d		4406 ^d	
			Multi-point	Two-point ^e	Multi-point	Two-point ^e
rs1037973	79,957,577	106.52	-	-	-8.15	-1.99
rs2012502	80,285,582	109.31	-	-	-5.25	-3.60

^aMarkers in italics denote marker limits for the locus based on the overlap of families 4338 and 4406 using the 3-unit support intervals and homozygous regions

^bPhysical position in base pair based on Build 36.1 of the human reference sequence

^cGenetic position in centimorgan from Rutgers combined linkage-physical map of the human genome Build 36 version

^dLOD scores in italics denote scores for limiting markers for homozygous regions

^eTwo-point LOD scores shown at $\theta = 0.0$