

## ABSTRACT

### CHARACTERIZATION OF A DFNB1 DELETION ALLELE IN A MICHIGAN KINDRED OF GERMAN DESCENT

By

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A novel DFNB1 allele identified in a kindred segregating recessively-inherited hearing loss contains a 131.4 kb deletion, upstream of the transcriptional start sites of both GJB2 and GJB6, the genes encoding the gap junction proteins connexin 26 and connexin 30, respectively. The four family members who are compound heterozygotes for this deletion and for the 35delG mutation of GJB2 are all profoundly deaf, and have been since infancy. Heterozygous carriers of this deletion have normal hearing. We demonstrate that this allele segregates with a low-expression phenotype of GJB2 and GJB6 mRNA, assayed by allele-specific PCR of cDNA made from buccal cell RNA of four individuals (three assayed for GJB2 expression, one assayed for GJB6 expression). Considering the evidence of this allele together with that of other DFNB1 deletion alleles, it is likely that the pathogenicity of the DFNB1 deletion alleles is a result of loss of one or more distal GJB2 enhancer elements. One or more critical GJB2 regulatory elements is likely to exist within the genomic interval that is common to the three DFNB1 deletions that leave the GJB2 sequence intact; this interval extends from about chr1 3:19,837,300-19,932,800 (Mar2006 assembly (NCBI36/hgl8)). Assays of candidate enhancer elements identified by cross-species conservation have shown that some sequences within the 131.4 kb deletion interval upregulate or downregulate transcription of a reporter gene in two human epithelial cell lines. Disruption of distal GJB2 regulatory elements may

underlie idiopathic hearing loss in persons with monoallelic mutation in the GJB2 coding region. As mutations in GJB2 are the most common cause of congenital hearing loss worldwide, understanding the c/s-regulatory landscape of this gene is important in assessing pathogenicity of extragenic sequence variants in suspected DFNB1 alleles, and in interpreting the variation in expressivity of hearing loss in persons with biallelic coding region mutations of GJB2.