Hearing Loss: **GJB2** Gene Sequencing

**Test Code:** OZ  
**Turnaround time:** 3 weeks  
**CPT Codes:** 81252 x1

### Condition Description

In the United States, approximately 1 in 1000 children are diagnosed with prelingual hearing loss (HL) or deafness. Approximately half of prelingual hearing loss or deafness is attributed to environmental exposures and the remaining half is attributed to genetic causes. Approximately 30% of hereditary hearing loss is estimated to be syndromic (associated with other birth defects) while the remaining 70% is non-syndromic (isolated and not associated with other findings). Non-syndromic deafness is mainly due to recessive genes (75-80%) and over 20 such genes have been identified, but non-syndromic deafness may also be inherited in autosomal dominant, X-linked, or mitochondrial patterns.

Molecular testing can aid in rapid diagnosis of hearing loss. Early diagnosis of hearing defects can provide diagnostic information, facilitate timely intervention, and assist with genetic counseling.

Connexins are transmembrane proteins that form channels that allow rapid transport of small molecules between cells; the proteins connexin 26 and connexin 30 interact to form a channel that functions in the inner ear. The **GJB2** gene encodes the connexin 26 protein and is involved in 50% of autosomal recessive hearing loss. The **GJB6** gene is located near the **GJB2** gene, and encodes the protein connexin 30. Patients with non-syndromic hearing loss have been found to have two mutations in connexin 26, two mutations in connexin 30, or compound heterozygosity for one mutation in connexin 26 and one mutation in connexin 30 [1,2].

This test involves sequencing of the entire coding sequence of the **GJB2** gene that encodes the connexin 26 protein.

### References:
- GeneReviews Clinical Summary

### Genes

**GJB2**

### Indications

This test is indicated for:

- Individuals with clinical findings consistent with non-syndromic hearing loss when mitochondrial etiologies have been ruled out and testing of connexin 30 has resulted in no mutations found or one mutation found.

### Methodology

PCR amplification of exons and flanking regions contained in the **GJB2** gene is performed on patient genomic DNA. Direct sequencing of amplification products is performed in both the forward and reverse directions using automated fluorescence dideoxy sequencing methods. Patient gene sequences are compared to a normal reference sequence. Sequence variations are then classified as mutations, benign variants unrelated to disease or variations of unknown clinical significance. Variants of unknown clinical significance may require further studies of the patient and/or family members.

This assay does not interrogate the promoter region, deep intronic regions, or other regulatory elements. Large deletions are not detected by this analysis.

### Detection

Mutations in the promoter region, some mutations in the introns, and other regulatory elements cannot be detected by this analysis. Large deletions and insertions will not be detected by this assay. Single and multiple exon deletions and duplications can be detected by the separate **GJB2** and **GJB6** deletion/duplication array. It is possible that some patients with a typical presentation may not carry a mutation detected by this analysis. This analysis may detect novel variants of unclear effect, which may require further studies.

### Specimen Requirements

**Submit only 1 of the following specimen types**

**Type:** Saliva

**Specify Requirements:**  
Oragene™ Saliva Collection Kit  
Oragene™ Saliva Collection Kit used according to manufacturer instructions. Please contact EGL for a Saliva Collection Kit for patients that cannot provide a blood sample.

**Specimen Collection and Shipping:**  
Please do not refrigerate or freeze saliva sample. Please store and ship at room temperature.

Disclaimer: This information is confidential and subject to change without notice. It may not be reproduced in whole or part unless authorized in writing by an authorized EGL representative.
Type: Whole Blood (EDTA)

Specimen Requirements:
EDTA (Purple Top)
Infants and Young Children (2 years of age to 10 years old): 3-5 ml
Older Children & Adults: 5-10 ml
Autopsy: 2-3 ml unclotted cord or cardiac blood

Specimen Collection and Shipping:
Ship sample at room temperature for receipt at EGL within 72 hours of collection. Do not freeze.

Type: DNA, Isolated

Specimen Requirements:
Microtainer
8µg
Isolation using the Perkin Elmer™ Chemagen™ Automated Extraction method or Qiagen™ Puregene kit for DNA extraction is recommended.

Specimen Collection and Shipping:
Refrigerate until time of shipment in 100 ng/µL in TE buffer. Ship sample at room temperature with overnight delivery.

Related Tests

- The Hearing Loss Panel (HL) is indicated for patients who have not have previous molecular testing and includes sequencing of the \textit{GJB2} and \textit{GJB6} genes, targeted mutation analysis of the \textit{GJB6} common 342kb deletion, and testing for mitochondrial mutations associated with aminoglycoside sensitivity.
- For patients with mutations not identified by full gene sequencing, a separate Deletion/Duplication Assay is available for connexin 26 and connexin 30 using a targeted CGH array. Refer to the test requisition or contact the laboratory for more information.
- Custom Diagnostic Mutation Analysis (KM) is available to family members if mutations are identified by sequencing.
- Prenatal testing is available to couples who are confirmed carriers of mutations. Please contact the laboratory genetic counselor to discuss appropriate testing prior to collecting a prenatal specimen.
- Mitochondrial Mutation Panel for patients with a history of aminoglycoside sensitivity (QJ)