ClinLab
Clinical Testing Laboratory, Inc.

Clinical Testing Lab of Washington
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| Specimen Number | Specimen Type | Control Number | Account Number | Account Phone Number | Route |
|---|---|---|---|---|---|
| | Peripheral Blood | | | | |

| Patient Last Name | Patient Barcode |
|---|---|
| | |

| Patient First Name | Patient Middle Name |
|---|---|
| Bo | |

| Patient SS# | Patient Phone | Total Volume |
|---|---|---|
| | | |

| Age (Y/M/D) | Date of Birth | Sex | Fasting |
|---|---|---|---|
| 10 y.o. | | Male | |

| Patient Address | Indication: Hemophilia, possibly type B<br>Family History: Family history unknown<br>Ethnicity: Possibly Han Chinese |
|---|---|
| | |

| Date and Time Collected | Date Entered | Date and Time Reported | Physician Name | NPI | Physician ID |
|---|---|---|---|---|---|
| | | | Jane Ferreiro, MD | | |

| Tests Ordered |
|---|
| Hemophilia Mutation Evaluation |

General Comments
**Please send a copy of the final report to the Molecular Science/M1 Training office via Fax at (202) 555-1212**

## Clinical test results for DNA Hemophilia Mutation Evaluation

| GENE | TEST RESULTS | EXPLANATION |
|---|---|---|
| F9 (Xq27.1) | p.Asp110Gly | **This result confirms the diagnosis of Hemophilia B**. This result should be interpreted in the context of clinical presentation and results of other laboratory tests (e.g., APTT, Factor IX Activity, Factor IX Inhibitor, etc.).<br>A PCR/sequencing study has detected one copy of the Asp110Gly (F9: g.15392A>G, c.329A>G or p.Asp110Gly) variation. The Asp110Gly variation is an A to G change at nucleotide position 15392 in the F9 gene. This encodes an amino acid at position 110 (glycine) that is different from the reference (aspartate) and may have implications on structure and or function of the resulting protein.<br>As males have only one copy of the X chromosome, a pathogenic variation in an X-linked gene renders the patient with only a mutated form of the gene. Thus, they are highly susceptible to development of significant abnormal hemophiliac symptoms. |
| F8 (Xq28) | Negative | |

### INDICATIONS FOR TESTING
Individuals with a diagnosis of hemophilia B, appropriate at-risk female relatives of probands with identified mutations, and hemophilia B carriers with genetic counseling, are candidates for testing.

### METHODOLOGY
Factor IX sequencing: All coding exons (1-8) and associated intron junctions of the Factor IX gene are analyzed by direct DNA sequence analysis using an automated fluorescent sequencing machine. When a mutation is detected, confirmation is carried out on an independent amplification of PCR using a second prep (B-prep) by sequencing in the opposite direction. If no mutation is found, sequence analysis is performed in both directions.

## PERFORMANCE

Factor IX sequencing: From previous experience, we have been able to detect factor IX gene mutations in about 99% of individuals with the diagnosis of hemophilia B with specificity of mutation detection in probands and carrier detection is also estimated to be greater than 99%.

## LIMITATIONS

The sequence analysis will not detect mutations located in regions of the Factor IX gene that are not analyzed (non-coding exon regions, intron regions other than the splice junctions, and upstream and downstream regions). The sequencing method also will not detect gross genetic alterations including most duplications, inversions, or deletions (in females). Some sequence alterations that may be detected (such as those causing missense or synonymous changes) will be of unknown clinical significance.

## CLINICAL DESCRIPTION

Hemophilia B is characterized by deficiency in factor IX clotting activity that results in prolonged oozing after injuries, tooth extractions, or surgery, and delayed or recurrent bleeding prior to complete wound healing. This is an X-linked recessive bleeding disorder with an incidence of about 1 per 30,000 live male births. Hemophilia B affects males, however, all male offspring will be normal, and although all female offspring will be obligatory carriers, they rarely have symptomatic bleeding. In contrast, female offspring of carriers of hemophilia B have a 50% chance of being carriers themselves, and each male offspring has a 50% chance of being affected.

The age of diagnosis and frequency of bleeding episodes are related to the level of factor IX clotting activity. In severe hemophilia B, spontaneous joint or deep-muscle bleeding is the most frequent symptom. Individuals with severe hemophilia B are usually diagnosed during the first two years of life; without prophylactic treatment, they may average up to two to five spontaneous bleeding episodes each month. Individuals with moderate hemophilia B seldom have spontaneous bleeding; however, they do have prolonged or delayed oozing after relatively minor trauma and are usually diagnosed before age five to six years; the frequency of bleeding episodes varies from once a month to once a year. Individuals with mild hemophilia B do not have spontaneous bleeding episodes; however, without pre- and post-operative treatment, abnormal bleeding occurs with surgery or tooth extractions; the frequency of bleeding may vary from once a year to once every ten years. Individuals with mild hemophilia B are often not diagnosed until later in life. In any individual with hemophilia B, bleeding episodes may be more frequent in childhood and adolescence than in adulthood. Approximately 10% of carrier females are at risk for bleeding (even if the affected family member has mild hemophilia B) and are thus symptomatic carriers, although symptoms are usually mild. After major trauma or invasive procedures, prolonged or excessive bleeding usually occurs, regardless of severity.

## REFERENCES


1. Yoshitake S, Schach BG, Foster DC, et al: Nucleotide sequence of the gene for human factor IX (antihemophilic factor B). Biochemistry 1985 July 2;24(14):3736-3750
2. Giannelli F, Green PM, Sommer SS, et al: Haemophilia B: database of point mutations and short additions and deletions-eighth edition. Nucleic Acids Res 1998 Jan 1;26(1):265-268
3. Ketterling RP, Bottema CD, Phillips JA 3rd, et al: Evidence that descendants of three founders constitute about 25% of hemophilia B in the United States. Genomics 1991 Aug;10(4):1093-1096