BLOODCENTER of WISCONSIN™

SEVERE CONGENITAL NEUTROPENIA AND CYCLIC NEUTROPENIA SEQUENCE ANALYSIS

BloodCenter of Wisconsin offers DNA sequencing of the ELANE and HAX1 genes for diagnosis of congenital and cyclic neutropenia.

BACKGROUND:

Severe congenital neutropenia (SCN) is a disorder of neutrophil production that is characterized by recurrent fever, infections and inflammation of the mouth, skin and pharynx. Predisposition to myelodysplastic syndrome and acute myeloid leukemia is also associated with SCN. Diagnosis is based on clinical findings and serial measurement of the absolute neutrophil count (ANC).[1,2] Cyclic neutropenia is distinguished from SCN by regular oscillations of the ANC, generally milder infectious complications, and no associated risk of malignancy.[2,3] Both SCN and cyclic neutropenia can be treated with granulocyte colony-stimulating factor.

Mutations in the *ELANE* gene, which codes for the neutrophil elastase protein, have been reported in 38-80% of SCN patients and in 90-100% of cyclic neutropenia patients.[2] *ELANE*-related neutropenia is inherited in an autosomal dominant manner and *de novo* mutations have been found in children with unaffected parents.[2,4] An autosomal recessive form of SCN (Kostmann Disease) is caused by mutations in the *HAX1* gene, which encodes a mitochondrial membrane protein.[5] Unpublished data from the European SCN International Registry suggests that *HAX1* mutations account for 15-20% of SCN cases.[6] Pathogenic heterozygous mutations in *ELANE* confirm the clinical diagnosis of either cyclic neutropenia or SCN. Although some *ELANE* variants have been associated with only one or the other diagnosis, other variants have been associated with both phenotypes.[2,7] In the absence of an *ELANE* mutation, identification of pathogenic homozygous mutations in the *HAX1* gene confirms a clinical diagnosis of the Kostmann Disease form of SCN.

REASONS FOR REFERRAL:

- Confirmation of diagnosis
- Evaluation of family members
- Prenatal diagnosis

METHOD:

PCR amplification and bidirectional DNA sequence analysis are performed. The complete coding region and splice junction of each exon is compared to the reference sequence (available from the lab on request).

LIMITATIONS:

Analytical sensitivity is >99%. Rare polymorphisms within primer or probe regions may interfere with detection of gene variants. Mutations that are outside the regions sequenced (e.g. regulatory regions) and large deletions or duplications will not be detected. Clinical sensitivity will be highest in patients presenting with the classic symptoms and the expected inheritance pattern. Other genes (*GFI1*, *WAS*, *CSF3R* and *G6PC3*) have been linked to SCN and cyclic neutropenia diagnoses in rare cases, and additional genes may also have yet undiscovered roles.[8] As a result of these limitations, a negative result does not exclude a possible genetic basis of the patient's phenotype.

PO Box 2178
Milwaukee, WI 53201-2178
Location/Sample Deliveries:
638 N. 18th St. Milwaukee, WI 53233-2121
p800-245-3117 | f414-937-6202 | www.bcw.edu

REPORTING OF RESULTS:
Results are reported in accordance with ACMG next-generation sequencing standards.[9]

- All *ELANE* and *HAX1* variants predicted to be pathogenic, likely pathogenic, and of unknown significance will be reported.
- Variants classified as likely benign or benign will not be reported but are available upon request.

SPECIMEN REQUIREMENTS:
7-15 ml amniotic fluid, $5x10^6$ cultured amniocytes, or 3-5 ml EDTA (lavender top) whole blood. Contact the laboratory to discuss prenatal sample requirements.

TURNAROUND TIME: 14 days

SHIPPING REQUIREMENTS:
Ship on an ice pack or at room temperature. Place the specimen and the requisition into plastic bags and seal. Insert into a Styrofoam container, seal and place into a sturdy cardboard box, and tape securely. Ship the package in compliance with your overnight carrier guidelines. Label with the following address:

Client Services/Platelet Neutrophil Immunology Laboratory
BloodCenter of Wisconsin
638 N. 18th St.
Milwaukee, WI 53233
Phone: 800-245-3117, ext. 6250

CPT CODES:
ELANE Sequence Analysis
CPT Codes: 81479

HAX1 Sequence Analysis
CPT Codes: 81479

REFERENCES:

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