Fanconi Anemia Gene Sequencing Panel

Genes Tested:

BRCA1	*BRCA2*	*BRIP1*	*ERCC4*
FANCA	*FANCB*	*FANCC*	*FANCD2*
FANCE	*FANCF*	*FANCG*	*FANCI*
FANCL	*FANCM*	*MAD2L2*	*PALB2*
RAD51	*RAD51C*	*RFWD3*	*SLX4*
UBE2T	*XRCC2*		

Disorder:

Fanconi Anemia (FA) is a rare, inherited chromosome-instability syndrome, estimated to occur in 1:100,000 live births. However, its prevalence is much higher in some populations including Ashkenazi Jewish, Spanish Gypsy, and black South African. A unique characteristic that distinguishes FA from other chromosome breakage syndromes is the cellular hypersensitivity to DNA cross-linking agents causing chromosome breakage. Much of the function of the FA proteins in normal cells is unclear. Spontaneous reversion of a pathogenic allele to wild type (or correction of cellular defect with a second variant) in patients who are homozygous for FA variants has been reported.

Patients with FA have varied clinical manifestations, most commonly progressive bone marrow failure at a median age of five (5) years, progressive pancytopenia and congenital malformations including short stature, radial aplasia, urinary tract abnormalities, hyperpigmentation, and/or developmental delay. FA patients also have a predisposition to cancer, particularly acute myeloid leukemia, as well as an increased risk of developing solid tumors in the head, neck, skin, GI tract, and genital tract. The symptoms of FA are highly variable, even among individuals within the same family or among patients within the same complementation group. Treatment depends on the symptoms experienced by the individual patient.

Pathogenic variants which result in premature termination of the *FANCB* protein are typically associated with a severe VACTERL-H phenotype which includes ventriculomegaly/hydrocephalus, radial ray defects with aplastic or hypoplastic thumbs, urinary tract abnormalities, vertebral defects, hypogonadism, gastrointestinal atresia and pre- or postnatal growth retardation, in addition to abnormal chromosome breakage and development of anemia.

Genetics:

FA is usually inherited as an autosomal recessive condition. However, Fanconi Anemia type B *(FANCB)* shows X-linked inheritance, and Fanconi Anemia type R (*RAD51*) shows autosomal dominant inheritance. Fanconi anemia is genetically heterogeneous and variants in several genes have been identified. However, pathogenic variants in three genes (*FANCA*, *FANCC*, and *FANCG*) account for disease in the majority of patients. Heterozygous carriers of inactivating mutations in *BRCA1, BRCA2, BRIP1, PALB2, RAD51, RAD51C* and *SLX4* are at some increased risk of developing breast and/or ovarian cancers.

Indications:

FA Panel by NGS:

- Confirmation of genetic diagnosis in a patient with a clinical diagnosis of Fanconi anemia
- Carrier identification in individuals with a family history of FA of unknown genetic basis.

Gene Specific Sequencing:

- Confirmation of genetic diagnosis in a patient with FA and in whom complementation studies suggest a specific genetic diagnosis.

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Variant Specific Analysis:

- Presymptomatic testing of at-risk relatives for medical management and prior to bone marrow donation
- Carrier identification in individuals in whom specific variant(s) have been identified in the proband with FA
- Prenatal diagnosis of an at-risk fetus, after confirmation of variant(s) in the parent(s) and by prior arrangement only.

Specimen:

At least 3 mLs whole blood in a lavender top (EDTA) tube or saliva in an Oragene saliva kit. Please call 513-636-4474 for a free saliva collection kit.

Note: For post-transplant patients, we accept pre-transplant samples or post-transplant skin fibroblasts ONLY (blood, saliva, and cytobrushes are not accepted). Culturing of skin fibroblasts is done at an additional charge.

Testing Methodology:

FA Panel by NGS: This test is performed by enrichment of the coding exons, flanking intronic and untranslated regions (5′ and 3′), as well as known pathogenic variants (HGMD 2018.4) in the promoter and deep intronic regions of the genes specified above using oligonucleotide probe hybridization followed by next-generation sequencing with >50X coverage at every target base. All pathogenic and likely pathogenic variants, as well as variants of unknown (indeterminate) significance, as determined bioinformatically, are confirmed by Sanger sequencing. Regions with <50X will be filled in by Sanger sequencing. A detailed non-coding variant list is available upon request.

Gene Specific Sequencing: PCR-based sequencing of the entire coding region and intron/exon boundaries of the specific gene.

Variant Specific Analysis: Sanger sequencing following PCR amplification of the targeted variant(s) of the specified gene of the specified gene and selected known pathogenic variants in the promotor and deep intronic regions.

Test Sensitivity:

Clinical Sensitivity: DNA sequencing detects >90% of reported pathogenic variants in *BRCA2, BRIP1, FANCE,* and *FANCG*. DNA sequencing detects ~60% of reported pathogenic variants in *FANCA*. The clinical sensitivity of the remaining genes on this panel is unknown. Large exonic deletions are common in *FANCA* and have been reported in some of the other genes on this panel. Deletion/duplication analysis may be indicated as a follow-up test in symptomatic patients with a normal sequencing result or a single (heterozygous) variant in one of the genes associated with autosomal recessive FA.

Analytical Sensitivity: The sensitivity of DNA sequencing is over 99% for the detection of nucleotide base changes, small deletions and insertions in the regions analyzed.

Limitations: Variants in regulatory regions and non-reported variants in untranslated regions may not be detected by this test. Large deletions/ duplications, large insertions and other complex genetic events will not be identified using sequencing methodology.

Note: Single gene sequencing and targeted variant analysis is also available for all genes on the Fanconi Anemia Panel. Deletion/duplication is available for many of the genes on this panel. For further details, visit: www.cincinnatichildrens.org/deldup.

Turn-Around Time:

- Fanconi Anemia panel: up to 6 weeks
- Single gene sequencing: up to 28 days

CPT Codes:

- **Fanconi Anemia Panel by NGS:** 81443
- **Single gene sequencing, targeted variant analysis, and deletion/duplication:** call for information.

Please call 1-866-450-4198 for current pricing, insurance preauthorization or with any billing questions.

Results:

Results will be reported to the referring physician or health care provider as specified on the requisition form.

Shipping Instructions:

Please enclose **test requisition** with sample.
All information must be completed before sample can be processed.

Place samples in styrofoam mailer and ship at room temperature by overnight Federal Express to arrive Monday through Friday.

Ship to:

Cytogenetics and Molecular Genetics Laboratories
3333 Burnet Avenue NRB 1042
Cincinnati, OH 45229
513-636-4474

References:

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