CENTER FOR PRECISION DIAGNOSTICS

Breast Cancer Solid Tumor Sequencing Panel

BRCA1	*BRCA2*	*PIK3CA*							

Indication

BReast CAncer genes 1 and 2 (*BRCA1* and *BRCA2*) are tumor-suppressor genes and their protein products have multiple functions, including serving as key enzymes in the homologous recombination pathway, which is the high-fidelity mechanism for repair of DNA double-strand breaks. Inactivation of either of these genes through germline or somatic loss of function mutations fundamentally influences cancer risk and development. Assessment of *BRCA1* and *BRCA2* mutation status in breast tumors is important since presence of germline or somatic mutations in *BRCA1* or *BRCA2* is predictive for treatment response to poly(ADP-ribose) polymerase (PARP) inhibitors and platinum agents. In a population without strong germline founder mutations, deleterious mutations in BRCA1 and BRCA2 were identified in 11% of breast carcinoma cases, with approximately 1/3 of these mutations being established as somatic and 2/3 as germline. Breast tumor phenotypes were also reported to be very similar regardless of the mutation being germline or somatic. The PARP inhibitors olaparib and talazoparib are FDA-approved for the treatment of patients with HER2-negative, metastatic breast cancer with deleterious or suspected deleterious germline *BRCA1* or *BRCA2* mutations. There is also promising clinical activity with olaparib in breast cancer patients with somatic mutations in BRCA1/2.

Phosphatidyl 3-kinases (PI3K) are a family of lipid kinases involved in many cellular processes, including cell growth, proliferation, differentiation, motility, and survival. *PIK3CA* (phosphatidylinositol-4,5-bisphosphate 3-kinase, catalytic subunit alpha) is the catalytic subunit of PI3-kinase, which is frequently mutated in a diverse range of cancers including breast, endometrial and cervical cancers. *PIK3CA* is altered in up to 30-35% of breast cancers. The alpha-selective PI3-kinase inhibitor alpelisib in combination with the Estrogen Receptor (ER)-antagonist fulvestrant is FDA-approved for the treatment of patients with *PIK3CA*-mutant ER+/HER2- breast cancer.

Testing method

This assay detects hotspot mutations and copy number alterations in multiple cancer-related genes relevant to clinical management and treatment of breast cancer. For each gene included on the clinical panel (listed below), the target exons are enriched by hybrid capture method followed by next generation sequencing (NGS) on the Illumina MiSeq instrument. This method was optimized for use with low quantity of input DNA (50 ng) obtained from formalin-fixed, paraffin-embedded (FFPE) tissues providing high on-target coverage with coverage uniformity above 95% throughout the entire target region. This analysis is performed on genomic DNA isolated from FFPE tumor tissue and does not differentiate between germline and somatic mutations. Data analysis is performed using SOPHiA DDM platform, which provides sequence alignment to reference genome, variant detection and annotation, interpretation of clinically significant genomic alterations and their association to approved or investigational therapies.

Diagnostic sensitivity: This assay is designed to detect known single nucleotide variants, insertions, deletions, and copy number alterations only within defined target regions. Large insertions and deletions that include genomic sequence outside of the defined target regions may not be detected. Gene rearrangements are not detected.

Technical sensitivity: This assay may not detect certain mutations if the proportion of tumor cells in the sample studied is less than 20%. Sensitivity for detection of copy number variants is reduced in samples with tumor fraction below 50%.

Clinical Panel

Gene	Exon / Amino Acid (AA) Coverage	Annotation Transcript
BRCA1	Full coding sequence	NM_007294
BRCA2	Full coding sequence	NM_000059
PIK3CA	Exons 2, 3, 6, 8, 10, 21	NM_006218

Genes Targeted for Copy Number Analysis

BRCA1, BRCA2, PIK3CA

Turnaround time

5-10 business days

Sample requirements

The presence of adequate tumor in the material submitted for analysis should be confirmed by a surgical pathologist. A section from archival paraffin material or frozen surgical biopsies should be confirmed to contain > 50% tumor by a surgical pathologist. If the submitted material for analysis contains < 50% of tumor, areas of predominant tumor will be microdissected, if possible, to enrich for neoplastic cells.

- Formalin-fixed, paraffin-embedded tissue
- 5-6 tissue sections (please include H&E slide and a copy of pathology report)
- Cytology slides

CPT codes

81162, 81309, G0452 (88363 or 88381 may apply)

References

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Ship Specimens to:

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