OncoGeneDx: Breast Cancer Surgical Panel

Panel Gene List: BRCA1, BRCA2, PALB2

Clinical Features:
In the general population, approximately 1 in 8 women (12%) will develop breast cancer in their lifetime (SEER). Most cases of breast cancer develop sporadically with no family history of the cancer; however, 5-10% of cases are thought to be due to a hereditary predisposition. The features suggestive of a hereditary cancer predisposition include: young age at diagnosis (before age 50), multiple primary cancers in a single individual, diagnosis of a cancer type that is not common in the general population (such as ovarian cancer, male breast cancer, or pancreatic cancer), and several relatives affected with related cancers spanning multiple generations.

The OncoGeneDx Breast Cancer Surgical Panel includes three genes associated with an increased level of breast cancer risk that may influence surgical management. These genes include BRCA1 and BRCA2, which are associated with hereditary breast and ovarian cancer syndrome and PALB2, which is associated with an increased risk for both breast and pancreatic cancer.

It is estimated that 20-25% of familial breast cancer risk can be attributed to pathogenic variants in the BRCA1 and BRCA2 genes (Easton 1999, Pharoah 2002, van der Groep 2011). The contribution of pathogenic variants in the PALB2 gene to familial breast cancer risk overall is less well-characterized but is likely lower than the contribution of BRCA1 and BRCA2 pathogenic variants.

Hereditary Breast and Ovarian Cancer syndrome (BRCA1 and BRCA2): Women with pathogenic variants in BRCA1 or BRCA2 have a 41-87% lifetime risk to develop breast cancer and an up to 63% risk for contralateral breast cancer (Antoniou 2003, Chen 2007, Claus 1996, Ford 1998, King 2003, Graeser 2009, Risch 2006). Studies have shown that the lifetime risk to develop ovarian cancer is between 24-54% for BRCA1 pathogenic variant carriers and 11-27% for BRCA2 pathogenic variant carriers (Antoniou 2003, Chen 2007, Ford 1998, King 2003, Risch 2006). Other cancers associated with pathogenic variants in BRCA1 and BRCA2 in women include fallopian tube carcinoma, primary peritoneal carcinoma, and uterine serous carcinoma (Levine 2003, Biron-Shental 2006, Pennington 2013). The lifetime risk for breast cancer in male BRCA1/2 pathogenic variant carriers is approximately 7% with a pathogenic variant in BRCA2 and slightly increased with a pathogenic variant in BRCA1 (Liede 2004, Tai 2007). Other malignancies reported in families with pathogenic variants in BRCA1 or BRCA2 include prostate cancer in men, as well as pancreatic cancer and melanoma in both men and women.
**PALB2**: Women with a pathogenic variant in **PALB2** have been estimated to have a 2 to 3-fold increased risk of breast cancer over the general population (Erkko 2008, Rahman 2007) resulting in a lifetime risk of approximately 25% to 40%. More recent data has suggested a lifetime risk (up to age 70) ranging from 33% to 58% depending on the individual’s family history of breast cancer (Antoniou 2014). Women with a pathogenic variant in **PALB2** who have a family history of early-onset breast cancer may have a lifetime risk up to 58% (Byrnes 2008, Antoniou 2014). Casadei et al. (2011) found that **PALB2** pathogenic variant carriers are 6-fold more likely to have a family history of pancreatic cancer, 1.3-fold more likely to have a family history of ovarian cancer and 4-fold more likely to have a family history of male breast cancer. Although the association of pathogenic variants in **PALB2** and pancreatic cancer has been established, the exact risks are not yet well-understood (Jones 2009, Slater 2010).

**Inheritance Pattern:**
All of the genes on this panel are associated with an autosomal dominant cancer risk. **BRCA2** and **PALB2** are also associated with an extremely rare condition when inherited in an autosomal recessive fashion. The specifics of this inheritance are outlined in the table below.

**Test Methods:**
Genomic DNA from the submitted specimen is enriched for the complete coding region and splice site junctions of the genes on the panel using a proprietary targeted capture system developed by GeneDx. The products are sequenced on an Illumina HiSeq instrument with 2x100 paired-end reads. The sequence is aligned to reference sequences based on human genome build GRCh37/UCSC hg19. Capillary sequencing is used to confirm all variants with clinical or uncertain significance and to analyze regions with inadequate coverage by Next Generation sequencing (NGS). If present, apparently homozygous variants are confirmed using alternate primer pairs to significantly reduce the possibility of allele drop-out. All sequence alterations are described according to the Human Genome Variation Society (HGVS) nomenclature guidelines. Concurrent deletion/duplication analysis from NGS data is performed for all relevant genes on the panel to detect multi-exonic and most single-exon deletions and duplications. For specimens with insufficient copy number data and for confirmation of identified copy number changes, exon-level array CGH, MLPA or other appropriate methods are used. Copy-number alterations are reported according to the International System for Human Cytogenetic Nomenclature (ISCN) guidelines. Benign and likely benign variants, if present, are not reported but are available upon request. Data analysis is performed using gene-specific filtering; the genes evaluated by this test are listed on the first page of the report.
Test Sensitivity:
The clinical sensitivity of sequencing and deletion/duplication analysis of the 3 genes included in the OncoGeneDx Breast Cancer Surgical Panel depends in part on the patient’s clinical phenotype and family history. In general, the sensitivity is highest for individuals with features suggestive of hereditary predisposition to cancer as outlined above. DNA sequencing will detect nucleotide substitutions and small insertions and deletions, while NGS-CNV analysis, array CGH, or MLPA will detect exon-level deletions and duplications. These methods are expected to be greater than 99% sensitive in detecting pathogenic variants identifiable by sequencing or CNV technology. The likelihood of a false positive result is expected to be <1%.

Technical Limitations: Neither sequencing, exon-level array CGH nor MLPA can reliably detect mosaicism, and cannot detect chromosomal aberrations. Deletions involving more than 20bp and insertions involving more than 10bp are not reliably detected by the sequencing methodology, and deletions or duplications of less than 250bp are not reliably detected by NGS-CNV analysis or array CGH. Regions of certain genes have inherent sequence properties that yield suboptimal data, potentially impairing accuracy of the results. In the absence of mRNA/cDNA studies, we cannot completely exclude the possibility of undetectable clinically significant variants in certain regions of these genes. False negatives may also occur in the setting of bone marrow transplantation, recent blood transfusion, or suboptimal DNA quality. In individuals with active leukemia or lymphoma or with known chronic myeloid or lymphoid neoplasms (such as low grade MDS, CML, ET, P. vera, PMF, CLL), there is a possibility that testing of specimens containing leukocytes may detect an acquired somatic variant, resulting in a false positive result. In this situation, please contact one of our genetic counselors to discuss the utility of submitting an alternate specimen. The ability to detect genetic variants and naming conventions can differ among laboratories. Rare false negatives, therefore, may occur when testing for a specific variant identified at a laboratory other than GeneDx, if a positive control is not provided. Based on the specific array design and technology used, the reported coordinates of duplications and deletions at the exon or gene level can slightly differ among family members tested but, in general, relatives are expected to have the same copy number variant.

<table>
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<th>Gene</th>
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<td>BRCA1</td>
<td>BREAST CANCER TYPE 1 SUSCEPTIBILITY PROTEIN</td>
<td>AD</td>
<td>Hereditary Breast and Ovarian Cancer (HBOC) syndrome: breast, ovarian, pancreatic, prostate &amp; endometrial serous cancer</td>
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BRCA2

BREAST CANCER TYPE 2
SUSCEPTIBILITY PROTEIN

AD
Hereditary Breast and Ovarian Cancer (HBOC) syndrome: breast, ovarian, pancreatic, prostate, melanoma & endometrial serous cancer

AR
Fanconi Anemia

PALB2

PARTNER AND LOCALIZER
OF BRCA2

AD
Breast, pancreatic & ovarian cancer

AR
Fanconi anemia

Because of evolving and expanding phenotypes, this list of cancer/tumor types is not exhaustive. Gene-specific risk for some of the cancers and other features listed are not well-defined.

Abbreviations:
AD – Autosomal Dominant
AR – Autosomal Recessive
CGH – Comparative genomic hybridization
MLPA – Multiplex ligation-dependent probe amplification

References: