

OncoGeneDx: *BRCA1* and *BRCA2* Sequencing In Hereditary Breast and Ovarian Cancer (HBOC)

Gene List: *BRCA1*, *BRCA2*

Clinical Features:

In the general population, approximately 1 in 8 women (12%) will develop breast cancer in their lifetime, and 1 in 75 women (1.4%) will be diagnosed with ovarian cancer in their lifetime (SEER). Most cases of breast or ovarian cancers develop sporadically with no family history of the cancer. Individual risk factors and exposures, such as age, pregnancy history, menstrual history, benign breast disease, radiation exposure, and alcohol intake, are known to modify a woman's chance of developing these types of cancers. However, 5-10% of breast cancer cases and 15-20% of ovarian cancer cases are thought to be due to a hereditary predisposition. The features suggestive of a hereditary cancer predisposition include: young age at diagnosis, multiple primary cancers in a single individual, diagnosis of a cancer type that is not common in general population (such as ovarian cancer, male breast cancer, or pancreatic cancer), and several relatives affected with related cancers spanning multiple generations.

Pathogenic *BRCA1* and *BRCA2* variants increase the lifetime risk for breast and ovarian cancer significantly over the general population risk. The chances to develop breast cancer begin increasing when a woman is in her mid-20s (King 2003). Women with pathogenic *BRCA1* or *BRCA2* variants have between a 41-87% lifetime risk to develop breast cancer and up to a 63% risk for a contralateral breast cancer (Antoniou 2003, Chen 2007, Claus 1996, Ford 1998, Graeser 2009, King 2003, Risch 2006). This risk depends on the age at which the first breast cancer was detected (Graeser 2009). The lifetime risk for breast cancer in males with a pathogenic *BRCA2* variant is approximately 7%, and slightly increased for those with a pathogenic *BRCA1* variant (Liede 2004, Tai 2007).

The risk of ovarian cancer begins to increase in the mid-30s, but becomes most significant in the 50s and beyond. The lifetime risk to develop ovarian cancer is between 24-54% for pathogenic *BRCA1* variant carriers and 11-27% for pathogenic *BRCA2* variant carriers (Antoniou 2003, Chen 2007, Ford 1998, King 2003, Risch 2006). Other associated cancers in women include fallopian tube carcinoma, primary peritoneal carcinoma, and uterine serous carcinoma (Levine 2003, Biron-Shental 2006, Pennington 2013).

The risk for other malignancies has been reported in families with pathogenic variants in *BRCA1* or *BRCA2* including prostate cancer in men as well as pancreatic cancer and melanoma in both men and women. Male and female pathogenic *BRCA2* variant carriers are

estimated to have up to a 7% risk for pancreatic cancer while male carriers are estimated to have up to a 34% risk for prostate cancer (Ozcelik 1997, The Breast Cancer Linkage Consortium 1999). Male pathogenic *BRCA1* variant carriers have been shown to have a slightly increased risk for prostate cancer before age 65 while pancreatic cancer have been suggested to also be slightly increased in both men and women (Brose 2002, Leongamornlert 2012, Liede 2004, Thompson 2002).

Two pathogenic variants in the *BRCA2* gene, one in each copy of the gene (biallelic pathogenic variants), are associated with an extremely rare autosomal recessive syndrome called Fanconi anemia. This condition is characterized by an increased risk for malignancy in children including leukemia and certain solid tumors as well as physical abnormalities and bone marrow failure. Therefore, if both mother and father are carriers of a pathogenic *BRCA2* variant, each of their children would have a 25% chance to inherit both variants, a 50% chance to inherit one of the variants, and a 25% chance to inherit neither variant.

Inheritance Pattern:

BRCA1 and *BRCA2* are associated with an autosomal dominant cancer risk. *BRCA2* is also associated with Fanconi Anemia when inherited in an autosomal recessive fashion. The specifics of this inheritance are outlined above.

Test Methods:

Genomic DNA from the submitted specimen was enriched for the complete coding region and splice site junctions of the *BRCA1* and *BRCA2* genes using a proprietary targeted capture system developed by GeneDx for dried blood spot specimens and PCR amplification (TruSeq Custom Amplicon) for all other specimen types. The products were sequenced on either an Illumina MiSeq or HiSeq instrument with 2x150 or 2x100 paired-end reads, respectively. The sequence was aligned to reference sequences based on human genome build GRCh37/UCSC hg19. Capillary sequencing was used to confirm all variants with clinical or uncertain significance and to analyze regions with inadequate coverage by Next Generation sequencing. If present, apparently homozygous variants were 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. Benign and likely benign variants, if present, are not reported but are available upon request.

GeneDx offers *BRCA1* and *BRCA2* Deletion and Duplication analysis as an additional test. The *BRCA1* and *BRCA2* genes are also included on our OncoGeneDx next generation sequencing panels for hereditary cancer.

Test Sensitivity:

Regarding clinical sensitivity, it is estimated that approximately 20-25% of familial breast cancer risk and 75% of hereditary ovarian cancer risk is attributed to pathogenic variants in the *BRCA1* and *BRCA2* genes (Easton 1999, Pharoah 2002, van der Groep 2011, Walsh 2011). This test is expected to be greater than 99% sensitive in detecting variants identifiable by sequencing. The likelihood of a false positive result is expected to be <1%. This test will not detect large genomic deletions or duplications in the *BRCA1* or *BRCA2* genes, which account for approximately 6-12% of cases of Hereditary Breast and Ovarian Cancer syndrome (Judkins 2012, Walsh 2006). *BRCA1* and *BRCA2* Deletion and Duplication Analysis is available at GeneDx.

Technical Limitations: Sequencing cannot 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. Regions of certain genes have inherent sequence properties that yield suboptimal data, potentially impairing accuracy of the results. 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. Additionally, rare false negatives may occur when testing for a specific variant identified at a laboratory other than GeneDx if a positive control is not provided. The ability to detect genetic variants and naming conventions can differ among laboratories.

Gene	Protein	Inheritance	Disease Associations
<i>BRCA1</i>	BREAST CANCER TYPE 1 SUSCEPTIBILITY PROTEIN	AD	Hereditary Breast and Ovarian Cancer (HBOC) syndrome: Breast, Ovarian, Pancreatic, Prostate & Endometrial serous cancer
<i>BRCA2</i>	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

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:

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