CASE STUDY

Mutation-Specific Testing Sets a Family at Ease

Gender: Female

Age: 65 years

Location: Mumbai

Diagnosis: Ovarian cancer

Strand Test: Germline Cancer Test

Key Findings:

- Identification of a *BRCA1* variant with duplication of exons 4-6, in the proband
- Proband's eligibility for PARP inhibitor therapy established
- Proband's children were tested for the presence of the same variant, using MLPA
- Proband's children, two sons and a daughter, have **NOT** inherited the deleterious mutation, establishing their low risk for hereditary cancers

Introduction

Hereditary breast and ovarian cancer is a serious health concern in India. Estimates from several studies have shown that in India, amongst women, breast cancer has become the number one cause of cancer-related mortality (1,2). A particularly noticeable trend is the early occurrence of breast cancer in the Indian population. The early and high incidence of this cancer, amongst Indian women, strongly indicates that a lot of cases of breast cancer are caused by heritable mutations in genes that cause cancer.

Therefore, identification of a genetic predisposition for cancer, within members of a family, before the actual incidence of cancer, is vital. A recent case referred to Strand Life Sciences is a good illustration of this fact.

Patient Profile

Nayanika Das*, a 65-year-old school teacher, had been experiencing some unusual symptoms such as bloating and lack of appetite. She had led a fairly active life coupled with balanced nutrition. Digestive problems were not something she encountered regularly, and this left her worried. Gradually her list of symptoms grew to include tiredness and fatigue, changes in bowel habits and pressure in the pelvic region. She eventually consulted her physician who, in turn, referred her to a prominent oncologist in Mumbai.

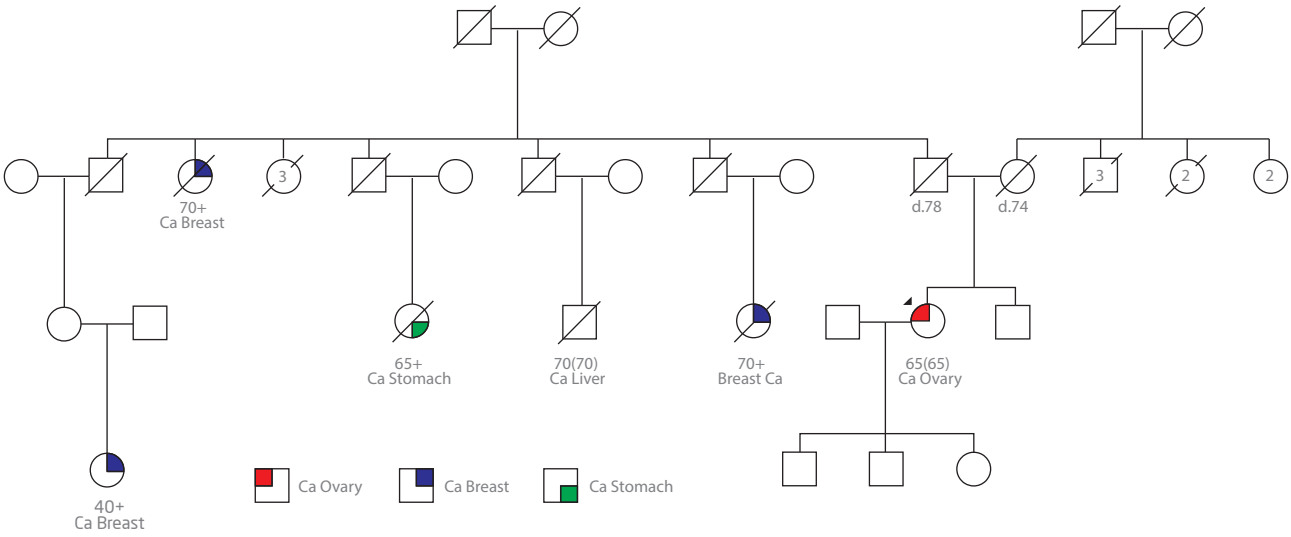
Nayanika's oncologist advised her a transvaginal ultrasound scan as well as a test to understand CA-125 protein levels in her blood. Since a mass was detected in the ovary, a biopsy was advised. Histopathological investigations of the tissue biopsy revealed that Nayanika was suffering from ovarian cancer.

Family Tree Pre-Genetic Testing

Nayanika's paternal cousin sister had been diagnosed with breast cancer at the age of 70 years and had succumbed to the disease. Another paternal cousin brother had suffered from stomach cancer, which proved fatal at the age of 65 years. Her paternal aunt had also been diagnosed with breast cancer at the age of 70 years and was lost to the disease.

** Name changed to protect patient privacy*

Given this family history, a paternal inheritance of genes that predispose a person to hereditary breast and ovarian cancer was suspected. Accordingly, Nayanika was advised to take the Strand Germline Cancer Test.



Treatment Options

Nayanika was advised chemotherapy for treatment of her ovarian cancer. Her oncologist also suggested a pedigree analysis to ascertain whether this was a case of familial/hereditary ovarian cancer.

Results of Genetic Testing

The Strand Germline Cancer Test was prescribed to Nayanika. This test is designed to identify heritable mutations in 19 genes that are associated with various hereditary cancer syndromes.

In Nayanika's case, a mutation in the *BRCA1* gene was found in her genome.

RESULT Positive for a heterozygous **'likely pathogenic'** variant, which causes heterzygous duplication of exons 4-6 in the *BRCA1* gene.

Key Findings

Gene	Variation	Zygosity	Clinical significance
BRCA1	chr14:41256139-?_41258550+?dup c:(134+1_135-1)_(441+1_442-1)dup Exon 4-6 duplication	Heterozygous	Likely Pathogenic

Exons 4-6 in the *BRCA1* gene were duplicated in her genome, resulting in loss of function of the resultant protein.

In this particular case, the identification of the duplication of a large fragment of the gene was enabled by StrandNGS-Strand's proprietary software for analysis of NGS data. Copy-number variants (CNVs) are otherwise difficult to detect by NGS techniques.

Mutation-Specific Testing of Other Family Members

Nayanika is a mother to two sons, Rakshit*, Kunal*, and a daughter, Abhilasha*. Since a hereditary *BRCA1* mutation was identified in her genome, she was worried about her children being carriers of the same mutation. *BRCA1* and *BRCA2* are the genes that when mutated and present even in heterozygous states, increase the risk to cause ovarian as well as breast cancer in people (3–5).

Rakshit, Kunal, and Abhilasha provided saliva samples for ascertaining their status as carriers of this *BRCA1* mutation. The MLPA (Multiplex Ligation Dependent Probe Amplification) technique was used to detect the same *BRCA1* variant in their DNA.

Results of MLPA testing of Rakshit's, Kunal's, and Abhilasha's DNA Samples

MLPA is a technique used to identify large deletions and duplications in a gene, in order to ascertain the number of copies in the gene of interest (6).

Gene name: *BRCA1* Genetic Alteration: chr17:41256139-?_41258550+?dup; c.(134+1_135-1)_(441+1_442-1)dup

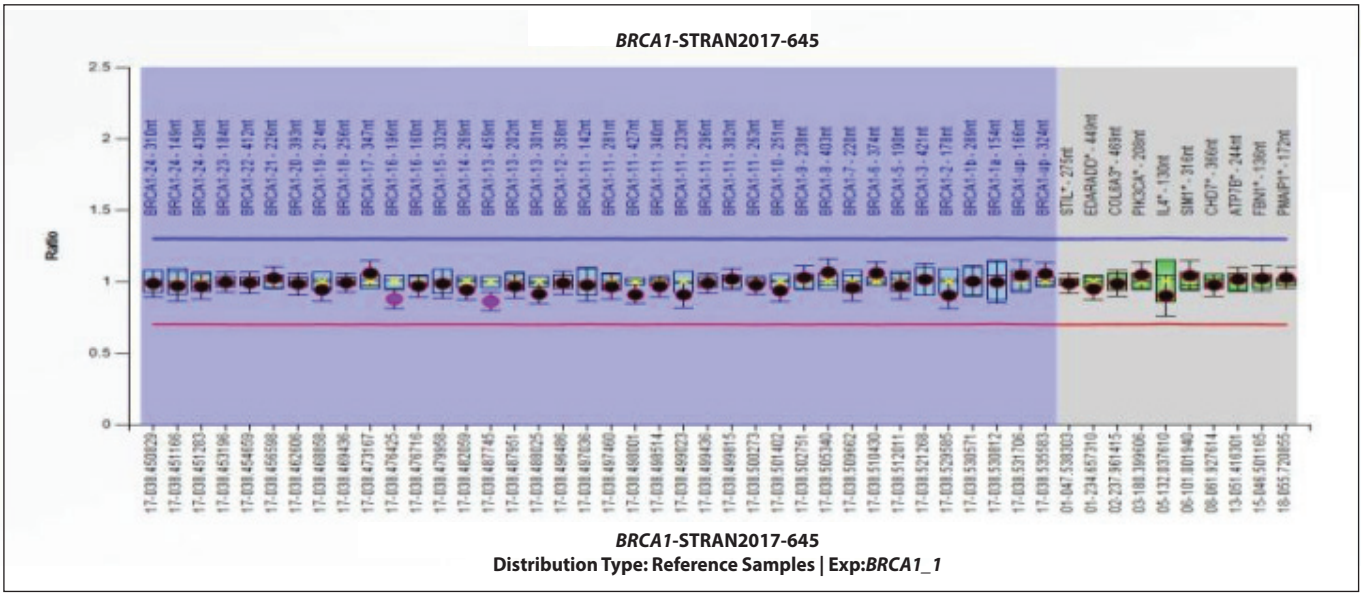


Figure 1: MLPA (multiplex ligation-dependent probe amplification) data from the individual showing normal copy of exons 4-6 of the *BRCA1* gene (Refseq id:NM_007294). This result was obtained from DNA samples from Rakshit, Kunal and Abhilasha.

In all these samples, the duplication of exons 4-6 was NOT detected. The MLPA testing confirmed that Rakshit, Kunal, and Abhilasha did not inherit the 'likely pathogenic' variant of the *BRCA1* gene that was detected in Nayanika's genome.

Conclusions

- A duplication variant (exons 4-6) in the *BRCA1* gene was identified in Nayanika Das' genome.
- This heritable (germline) variant was considered to be 'likely pathogenic' based on the fact that it leads to loss of function in the protein.
- The identification of a germline *BRCA1* mutation established Nayanika's eligibility to receive PARP inhibitor therapy (7–11). This targeted therapy has been shown to improve the quality of life in ovarian cancer patients, especially if they develop resistance to platinum-based chemotherapy drugs.
- Mutation-specific testing*, using the MLPA technique, helped to establish that Nayanika's children have NOT inherited this harmful mutation. They are not at enhanced risk for developing cancers due to this variant.
- Mutation-specific testing of relatives of index patients is a reliable method of ascertaining personal risk, as well as large-scale screening to understand the prevalence of germline mutations associated with hereditary cancer syndromes. This result was obtained from DNA samples from Rakshit, Kunal, and Abhilasha

The parameter marked with an * are not accredited by NABL and CAP.

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