BRCA1 & BRCA2 DELETION/DUPLICATION REPORT BY MLPA

Booking ID:
Name :
Sex/Age :
Referring Clinician: N/A

Sample Type:
Date & Time Collected:
Date & Time Received:
Date & Time Reported:

CLINICAL INDICATION

NA

RESULT SUMMARY

NEGATIVE
(No deletion/duplication detected)

Sr. No.	Deletion/Duplication	No. of Exons (Deleted/Duplicated)	MLPA probe ratio (Dosage quotient)	Clinical relevance
1	BRCA1 and BRCA2 gene	--	1.00	--

Data from Clinical sample:

Figure 1: MLPA Ratio chart: BRCA1

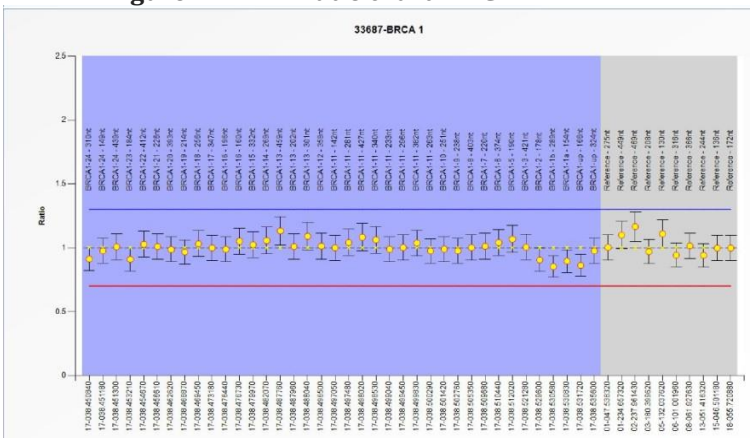
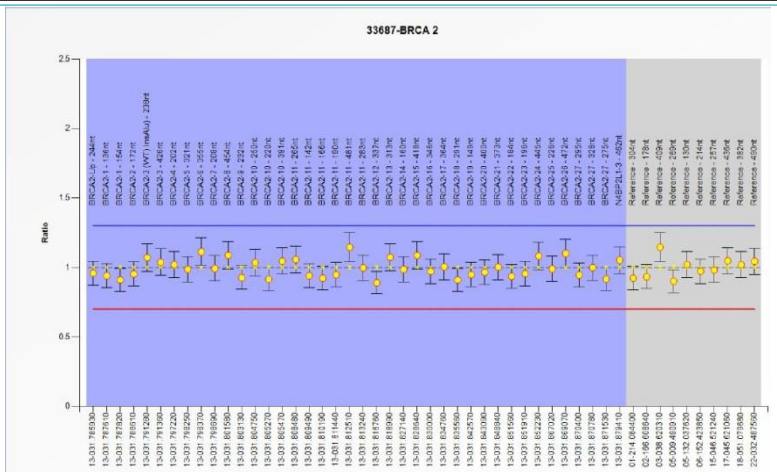


Figure 2: MLPA Ratio chart: BRCA2 Probemix



Interpretation Reference:	
Dosage Quotient [DQ] Distribution	**Copy Number Status**
DQ = 0	0 copies (homozygous deletion)
0.40 < DQ < 0.65	2 – 1 copy (heterozygous deletion)
0.80 < DQ < 1.20	NORMAL (identical to reference samples)
1.30 < DQ < 1.65	2 – 3 copies (heterozygous duplication)
1.75 < DQ < 2.15	2 – 4 copies (or 1 – 2copies)
MLPA ratios below 0.7 or above 1.3 indicate a heterozygous deletion or duplication respectively.	

TEST BACKGROUND

Breast and ovarian carcinomas are among the most common malignancies in developed countries. Most cases are considered sporadic, but in a substantial portion, a clear history of cases within a family is present. The BRCA1 and BRCA2 proteins are associated with the activation of double-strand break repair and homologous recombination and are important in maintaining genomic stability. Germline mutations in the BRCA1 and BRCA2 genes are linked to a high risk of young-onset hereditary breast and ovarian cancer. Features characteristic of hereditary, versus sporadic, breast cancer is younger age at diagnosis, frequent bilateral disease, and more frequent occurrence of disease among male relatives. Mutations in the BRCA1 and BRCA2 genes account for about 20 to 25% of hereditary breast cancers (Easton 1999) and about 5 to 10% of all breast cancers (Campeau et al. 2008). In addition, mutations in the BRCA1 and BRCA2 genes cause around 15% of ovarian cancers overall (Pal et al. 2005). Deletions or duplications are more frequent for BRCA1 than for BRCA2. The prevalence of deletions or duplications is dependent on the studied population and ranges from 0% to 11% of all BRCA1 and BRCA2 mutations.

TEST METHODOLOGY

THE MLPA TECHNOLOGY:

Multiplex ligation-dependent probe amplification (MLPA) is a variation of the multiplex polymerase chain reaction that permits multiple targets to be amplified with only a single primer pair. Each probe consists of two oligonucleotides which recognize adjacent target sites on the DNA. One probe oligonucleotide contains the sequence recognized by the forward primer, the other the sequence recognized by the reverse primer. Only when both probe oligonucleotides are hybridized to their respective targets, can they be ligated into a complete probe. The advantage of splitting the probe into two parts is that only the ligated oligonucleotides, but not the unbound probe oligonucleotides, are amplified.

Each complete probe has a unique length, so that its resulting amplicons can be separated and identified by (capillary) electrophoresis. Comparing the peak pattern obtained on a given sample with that obtained on various reference samples, the relative quantity of each amplicon can be determined. This ratio is a measure for the ratio in which the target sequence is present in the sample DNA.

BRCA1 and BRCA2 Deletion/Duplication Analysis is based on the MLPA technology (Multiplex Ligation-dependent Probe Amplification) and employs the SALSA® MLPA® probe mixes available from MRC (Holland) and entire analysis is done by Coffalyser software.

DISCLAIMER

- In most populations, the major cause of genetic defects in the BRCA1 and BRCA2 gene are small (point) mutations, most of which will not be detected by using SALSA MLPA Probemix BRCA1 and 2.
- MLPA cannot detect any changes that lie outside the target sequence of the probes and will not detect copy number neutral inversions or translocations. Even when MLPA did not detect any aberrations, the possibility remains that biological changes in that gene or chromosomal region do exist but remain undetected.
- Sequence changes (e.g., SNPs, point mutations, small indels) in the target sequence detected by a probe can cause false positive results. Mutations/SNPs (even when >20nt from the probe ligation site) can reduce the probe signal by preventing ligation of the probe oligonucleotides or by destabilizing the binding of a probe oligonucleotide to the sample DNA.
- For questions about this report, or for assistance in locating nearby genetic counselling services, please contact the Laboratory: contact@redcliffelabs.com.
- Although all precautions are taken during DNA tests the currently available data indicate that the technical error rate for all types of DNA analysis is approximately 2%. It is important that all clinicians or persons requesting DNA diagnostic tests are aware of these data before acting upon these results.

REFERENCES

- Schouten JP et al. (2002) Relative quantification of 40 nucleic acid sequences by multiplex ligation-dependent probe amplification Nucleic Acids Res 30, e57.
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- Hereditary breast cancer: new genetic developments, new therapeutic avenues. Hum Genet. 124:31-42. Casilli F et al. (2006).

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