Sanger Validation-Variant

PATIENT	REPORT DATE	BOOKING ID
Archita Singh	8 July 2022	#012206070082

Test Description
The MolQ Sanger Validation test confirms the variants obtained from high through put technology using sanger sequencing.

Clinician
Clinician Name: Dr Amit Verma
Medical Facility: Dr AV Institute of Personalized Cancer Therapy and Research
Pathologist: Not Provided

Patient Demographic
Name: Ms Archita Singh
Sex: Female
Date of Birth/Age: 46 years
Disease: Carcinoma Breast

Specimen
Booking ID: 012206070082
Site: NA
Sample Type: Blood
Date of Collection: 07-06-2022
Date of Booking: 07-06-2022

CLINICAL SYNOPSIS

Ms. Archita Singh, presented with clinical indications of Carcinoma Breast (right). The NGS sequencing data analysis has identified variant, c.1340+1G>A in *BRIP1* gene [Test Outside MolQ Laboratory]. The same variation is being validated by Sanger sequencing.

RESULTS

Variant is confirmed to be present by Sanger sequencing.

Analysis For: Variation in *BRIP1* Gene		Gene Name: *BRIP1* (Intron 9)
S.No.	**Variation Detected in NGS**	**Sanger Validation Result**
1.	chr17:59876460C>T; c.1340+1G>A (splice donor variant)	Present (Heterozygous)

*The variant analysis in Sanger sequencing is based on the *BRIP1* gene reference sequence NM_032043.3[1]. The intron number and nucleotide numbers will differ based on the reference file chosen and the database used.

CLINICAL CORRELATION AND VARIANT INTERPRETATION

Variant description: A heterozygous variant in intron 9 of the ***BRIP1*** gene (**chr17:59876460C>T; c.1340+1G>A**) was reported in Ms. Archita Singh by NGS.

The same variation was detected in heterozygous condition in this patient by Sanger sequencing (Figure 1).

Jatinder Kaur, PhD
Head, Molecular Biology & Genomics

Dr. Gulshan Yadav, MD
Head, Pathology

APPENDIX 1: TEST METHODOLOGY

METHOD

Targeted gene Sanger sequencing: Intron 9 of the *BRIP1* gene was PCR-amplified and the product was sequenced using Sanger sequencing. In case of mosaicism in leucocytes, the detection limits of Sanger sequencing for presence of variants are ~20%. The sequences were aligned to available reference sequences NM_032043.3[1] to detect variants using variant analysis software programs.

DISCLAIMER

About 0.44% of total cases are susceptible to allele dropout/dropin phenomenon, which can lead to misdiagnosis[2].

REFERENCES

1. https://www.ncbi.nlm.nih.gov/nuccore/NM_032043
2. Blais, Jonatan *et al.* Risk of Misdiagnosis Due to Allele Dropout and False-Positive PCR Artifacts in Molecular Diagnostics. The Journal of Molecular Diagnostics, Volume 17, Issue 5, 505 – 514.

Figure 1: Sequence chromatogram and alignment to the reference sequence showing the variant in intron 9 of the *BRIP1* gene [chr17:59876460C>T; c.1340+1G>A (splice donor variant)] detected in heterozygous condition in Ms. Archita Singh.