Accession ID:	**XXXXX**
Sample Type:	Plasma
Patient's Name:	**XXXXX**
Gender/Age:	Female/28 Years

CTDNA BREAST PANEL

Data Uploaded On:	xxxxxxx	**Report Generated On:**	xxxxxxx

CANCER TYPE:

Breast Cancer

RESULTS:

List of clinically significant mutations (CSM)

Variants of strong clinical significance:

GENE	VAF	FDA Approved Drugs		
		Responsive	Resistance	Total Clinical Evidence
BRCA2 (p.Ser3366*)	57.14%	Rucaparib		

Technical Information:

Variant	Depth	Genomic location	VAF
BRCA2(NM_000059.4);c.10095_10096insT	Total: 21 Alt: 12	chr13:32398608	57.14
Gene summary: Breast cancer 2, early onset (BRCA2) is a tumor suppressor gene that encodes a protein that functions in maintaining genomic stability, due to its involvement in the homologous recombination pathway for double-strand DNA repair. BRCA2 germline mutations including recurrent missense mutation N372H increase the risk of developing ovarian and/or breast cancer and somatic mutations are highest in colon, NSCLC, and ovarian cancers			

Therapeutic Summary:

Drug	Biomarker	Resistance/Responsive	Level	Clinical Indication
Rucaparib (Rubraca®)	BRCA2	**Responsive** | Supports	A	Peritoneum Cancer

Recommendations

- Genetic counseling is advised for interpretation on the consequences of the variant(s).
- If results obtained do not match the clinical findings, additional testing should be considered as per referring clinician's recommendation.
- Genetic test results are reported based on the recommendations of Association for Molecular Pathology, American Society of Clinical Oncology, and College of American Pathologists
- Sanger validation is recommended for the above listed variants.
- The significance/classification of the variant(s) may change based on genetic testing in the parents and other family members.
- Data revaluation performed upon the up gradation of databases used and results may vary in accordance.

AMP-ASCO-CAP CLASSIFICATION CRITERIA	Methodology
Four-tiered system to categorize somatic sequence variations based on their clinical significance **1. Tier I -** Variants of strong clinical significance. **2. Tier II -** Variants of potential clinical significance. **3. Tier III -** Variants of unknown clinical significance **4. Tier IV -** Benign or likely benign variants	The Genes2Me ctDNA breast NGS panel was used for sequencing that screens for 27 clinically relevant genes (coding regions of the genome) for diseases associated with genetic mutations. It covers all major mutations like SNV, InDels, adding up to a target size of 99Kb with hybridization-based target capture technique. After raw data generation, we follow the GATK best practices framework for identification of variants in the sample, starting with raw data quality check using the FastQC followed by BWA read aligner for mapping/aligning to human reference genome GRCh38. After the alignment, GATK Mutect2 algorithm is used for variant calling. Annotation of the variants is performed using open-source available software SnpEff. Further, clinically relevant mutations are annotated using published variants in literature and set of diseases databases – ClinVar, OMIM, COSMIC and HPO. The 1000Genome, gnomAD, dbSNP databases are used for annotation of variants for their minor allele frequency. The dbNSFP database is used for annotation and functional prediction of all Potential non-synonymous variants.

Limitations	Disclaimer
• Genetic testing is an important part of the diagnostic process however it may not always give a definitive answer. In some cases, a genetic variant may be missed due to the limitations in existing medical knowledge and testing technology. Accurate interpretation of test results is dependent on the availability of biological & medical information (clinical history) of the family, failing to this may leads to incorrect result interpretation and diagnosis. • Test results are interpreted in the context of clinical findings, available scientific evidences, family history and other laboratory data. The variation(s) which is/are potentially relevant – significant related to the patient's provided medical history is/are reported. • Genetic testing is highly accurate but rarely inaccurate results may occur for various reasons like mislabeling of samples, inaccurate clinical/medical family history, rare technical errors or unusual circumstances such as bone marrow transplantation, blood transfusion or the presence of change(s) in such a small percentage of cells that may not be detectable (mosaicism).	• The interpretation of variants in this report is performed to the best knowledge of the scientific & medical information available at the time of reporting. The classification of variants is based on AMP-ASCO-CAP guidelines. • As of the inherent technological limitations of the sequencing assay, some of the coding regions can't be properly sequenced, so, variations in these regions may not be identified & interpreted. It is possible that variants not identified by the assay may be associated with the provided phenotypes of the patient. • It is possible due to insufficient phenotypic information, a gene – variant may be present in data but not selected for analysis and interpretation. • The mutations have not been confirmed by Sanger sequencing. • Genes2Me clarify that the generated report(s) doesn't provide any kind of diagnosis or opinion or recommendation for any disease and its cure in any manner. It is therefore recommended that the patient and/or the guardian(s) of the patient must take the consultation of the clinician or a certified physician or doctor for further course of action. • If the provided material quality and/or quantity not up to the desired level, further procedures will be completed only after getting confirmation from referring clinician/physician only, so, in that case, test(s) result(s) may be misleading or even wrong, therefore, Genes2Me hereby disclaims all liability arising in this connection with the test(s) and report(s). • The analysis pipeline is developed in-house and the performance characteristics of this analysis are determined by Genes2Me only. • This test result should be used as a reference by the healthcare provider for diagnosis and development of treatment plan. • The clinically significant mutations enlisted in this report are provided as a professional service, and are not reviewed or approved by the FDA.

References

1. Richards S et al., ACMG Laboratory Quality Assurance Committee – Standards and guidelines for the interpretation of sequence variants: a joint consensus recommendation of the American College of Medical Genetics and Genomics and the Association for Molecular Pathology. Genet Med. 2015 May;17(5):405-24. 2.
2. Kalia SS et al., Recommendations for reporting of secondary findings in clinical exome and genome sequencing, 2016 update (ACMG SF v2.0): a policy statement of the American College of Medical Genetics and Genomics. Genet Med. 2017 Feb;19(2):249-255. 3.
3. Andrews S, FastQC: A Quality Control Tool for High Throughput Sequence Data, 2010, (http://www.bioinformatics.babraham.ac.uk/projects/fastqc/). 4.
4. Bolger AM et al., Trimmomatic: a flexible trimmer for Illumina sequence data. Bioinformatics (Oxford, England) (2014) Vol. 30,15: 2114-20. 5.
5. Li H. et al., Fast and accurate long-read alignment with Burrows-Wheeler transform. Bioinformatics (2010), 26(5):589-95. 6.
6. Quinlan AR et al., BEDTools: a flexible suite of utilities for comparing genomic features. Bioinformatics (Oxford, England) (2010) Vol. 26,6: 841-2.
7. McKenna A et al., The Genome Analysis Toolkit: a MapReduce framework for analyzing next-generation DNA sequencing data. Genome research (2010) Vol. 20,9: 1297-303. 8.
8. Cingolani P et al., A program for annotating and predicting the effects of single nucleotide polymorphisms, SnpEff: SNPs in the genome of Drosophila melanogaster strain w1118; iso-2; iso-3. Fly (2012) Vol. 6,2: 80-92. 9.
9. Clarke L et al. The international Genome sample resource (IGSR): A worldwide collection of genome variation incorporating the 1000 Genomes Project data. Nucleic acids research (2017) Vol. 45,D1: D854-D859. 10.
10. Karczewski KJ et al., The mutational constraint spectrum quantified from variation in 141,456 humans. Nature (2020) Vol. 581,7809: 434-443. 11.
11. GenomeAsia100K Consortium. The GenomeAsia 100K Project enables genetic discoveries across Asia. Nature (2019) 576, 106–111. 12.
12. Landrum MJ et al., ClinVar: improving access to variant interpretations and supporting evidence. Nucleic acids research (2018) Vol. 46,D1: D1062-D1067. 13.
13. McKusick VA, Mendelian Inheritance in Man. A Catalog of Human Genes and Genetic Disorders. Baltimore: Johns Hopkins University Press (12th edition), 1998. 14.
14. Köhler S et al., The Human Phenotype Ontology in 2021. Nucleic acids research (2021) Vol. 49,D1: D1207-D1217. 15.
15. Sherry ST et al., dbSNP—Database for Single Nucleotide Polymorphisms and Other Classes of Minor Genetic Variation. Genome Res. (1999), 9, 677–679. 16.
16. Liu, X et al., dbNSFP v4: a comprehensive database of transcript-specific functional predictions and annotations for human nonsynonymous and splice-site SNVs. Genome Med (2020) Vol. 12, 103. 17.
17. Mur P, García-Mulero S, Del Valle J, Magraner-Pardo L, Vidal A, Pineda M, Cinnirella G, Martín-Ramos E, Pons T, López-Doriga A, Belhadj S, Feliubadaló L, Munoz-Torres PM, Navarro M, Grau E, Darder E, Llort G, Sanz J, Ramón Y Cajal T, Balmana J, Brunet J, Moreno V, Piulats JM, Matías-Guiu X, Sanz-Pamplona R, Aligué R, Capellá G, Lázaro C, Valle L. Role of POLE and POLD1 in familial cancer. Genet Med. 2020 Dec;22(12):2089-2100. doi: 10.1038/s41436-020-0922-2. Epub 2020 Aug 14. PMID: 32792570; PMCID: PMC7708298. 18.
18. Li MM, et al. Standards and Guidelines for the Interpretation and Reporting of Sequence Variants in Cancer: A Joint Consensus Recommendation of the Association for Molecular Pathology, American Society of Clinical Oncology, and College of American Pathologists. J Mol Diagn. 2017 Jan;19(1):4-23. doi: 10.1016/j.jmoldx.2016.10.002. PMID: 27993330; PMCID: PMC5707196. 19.
19. John G Tate et al., COSMIC: the Catalogue Of Somatic Mutations In Cancer, Nucleic Acids Research, Volume 47, Issue D1, 08 January 2019, Pages D941–D947
20. Franklin Link:- https://franklin.genoox.com/clinical-db/home

***This is an electronically generated report, hence doesn't require any signature.**

***For any further technical queries please contact at contact@genes2me.com**