INVITAE

Name	DOB
Jane Doe	

Patient Name	DOB	Sex	MRN	Invitae #
Jane Doe		Female		

Clinical Team	Report Date	Sample Type	Sample Collection Date	Sample Accession Date
		Blood		

Test Performed
Sequence analysis and deletion/duplication testing of the 9 genes listed in the results section below.
- Add-on ATM Gene
- Add-on CHEK2 Gene
- Invitae Breast Cancer STAT Panel

Reason for Testing
Family history

Summary

Negative result. No Pathogenic sequence variants or deletions/duplications identified.

Clinical Sumary

- This negative test result does not eliminate the possibility that this individual's condition has a genetic component. Clinical follow up of this individual and their family members may still be warranted.
- These results should be interpreted within the context of additional laboratory results, family history, and clinical findings. Genetic counseling is recommended to discuss the implications of this result. For access to a network of genetic providers, please contact Invitae at clientservices@invitae.com, or visit www.nsgc.org or tagc.med.sc.edu/professional_organizations.asp.

Complete Results

The following genes were evaluated for sequence changes and exonic deletions/duplications:
ATM, BRCA1, BRCA2, CDH1, CHEK2, PALB2, PTEN, STK11, TP53

Results are negative unless otherwise indicated

Benign, Likely Benign, and silent and intronic variants with no evidence towards pathogenicity are not included in this report but are available upon request.

Methods

- Genomic DNA obtained from the submitted sample is enriched for targeted regions using a hybridization-based protocol, and sequenced using Illumina technology. Unless otherwise indicated, all targeted regions are sequenced with ≥50x depth or are supplemented with additional analysis. Reads are aligned to a reference sequence (GRCh37), and sequence changes are identified and interpreted in the context of a single clinically relevant transcript, indicated below. Enrichment and analysis focus on the coding sequence of the indicated transcripts, 10bp of flanking intronic sequence (20bp for BRCA1/2), and other specific genomic regions demonstrated to be causative of disease at the time of assay design. Promoters, untranslated regions, and other non-coding regions are not otherwise interrogated. For some genes only targeted loci are analyzed (indicated in the table above). Exonic deletions and duplications are called using an in-house algorithm that determines copy number at each target by comparing the read depth for each target in the proband sequence with both mean read-depth and read-depth distribution, obtained from a set of clinical samples. All clinically significant observations are confirmed by orthogonal technologies, except individually validated variants and variants previously confirmed in a first-degree relative. Confirmation technologies include any of the following: Sanger sequencing, Pacific Biosciences SMRT sequencing, MLPA, MLPA-seq, Array CGH. Array CGH confirmation of NGS CNV calling performed by Invitae Corporation (1400 16th Street, San Francisco, CA 94103, #05D2040778). The following analyses are performed if relevant to the requisition. For PMS2 exons 12-15, the reference genome has been modified to force all sequence reads derived from PMS2 and the PMS2CL pseudogene to align to PMS2, and variant calling algorithms are modified to support an expectation of 4 alleles. If a rare SNP or indel variant is identified by this method, both PMS2 and the PMS2CL pseudogene are amplified by long-range PCR and the location of the variant is determined by Sanger sequencing of the relevant exon in both long-range amplicons. If a CNV is identified, MLPA or MLPA-seq is run to confirm the variant. If confirmed, both PMS2 and PMS2CL are amplified by long-range PCR, and the identity of the fixed differences between PMS2 and PMS2CL are Sanger sequenced from the long-range amplicon to disambiguate the

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location of the CNV. Technical component of confirmatory sequencing is performed by Invitae Corporation (1400 16th Street, San Francisco, CA 94103, #05D2040778).

- The following transcripts were used in this analysis: ATM (NM_000051.3), BRCA1 (NM_007294.3), BRCA2 (NM_000059.3), CDH1 (NM_004360.3), CHEK2 (NM_007194.3), PALB2 (NM_024675.3), PTEN (NM_000314.4), STK11 (NM_000455.4), TP53 (NM_000546.5).
- A PMID is a unique identifier referring to a published, scientific paper. Search by PMID at http://www.ncbi.nlm.nih.gov/pubmed.
- An rsID is a unique identifier referring to a single genomic position, and is used to associate population frequency information with sequence changes at that position. Reported population frequencies are derived from a number of public sites that aggregate data from large-scale population sequencing projects, including ExAC (http://exac.broadinstitute.org) and dbSNP (http://ncbi.nlm.nih.gov/SNP).
- A MedGen ID is a unique identifier referring to an article in MedGen, NCBI's centralized database of information about genetic disorders and phenotypes. Search by MedGen ID at http://www.ncbi.nlm.nih.gov/medgen. An OMIM number is a unique identifier referring to a comprehensive entry in Online Mendelian Inheritance of Man (OMIM). Search by OMIM number at http://omim.org/.

Limitations

- This assay achieves >99% sensitivity and specificity for single nucleotide variants and insertions and deletions <15bp indels, based on validation study results. Sensitivity to detect insertions and deletions larger than 15bp but smaller than a full exon may be marginally reduced. Expansions and contractions within trinucleotide repeat regions may not be detected unless specified. Invitae's deletion/duplication analysis determines copy number with high confidence at >95% of targeted exons. This methodology may not detect low-level mosaicism. This report reflects the analysis of an extracted DNA sample. In very rare cases, (circulating hematolymphoid neoplasm, bone marrow transplant, recent blood transfusion) the analyzed DNA may not represent the patient's constitutional genome.

This report has been reviewed and approved by:

Placeholder for signature

Disclaimer

DNA studies do not constitute a definitive test for the selected condition(s) in all individuals. It should be realized that there are possible sources of error. Errors can result from trace contamination, rare technical errors, rare genetic variants that interfere with analysis, recent scientific developments, and alternative classification systems. This test should be one of many aspects used by the healthcare provider to help with a diagnosis and treatment plan, but it is not a diagnosis itself. This test was developed and its performance characteristics determined by Invitae. It has not been cleared or approved by the FDA. The laboratory is regulated under the Clinical Laboratory Improvement Act (CLIA) as qualified to perform high-complexity clinical tests (CLIA IDs: 05D2040778). This test is used for clinical purposes. It should not be regarded as investigational or for research.

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