Patient		Sample		Physician	
Name	**Jane Jones**	Specimen Type	**Blood**	Ordering Physician	**John Smith**
Date of Birth (Age)	**11/27/1940 (85 yrs)**	Collection Date	**08/09/2026**	Medical Facility	**BillionToOne Inc**
Assigned Sex at Birth	**Female**	Receipt Date	**08/10/2026**	Address	**1035 O'Brien Drive Menlo Park, California 94025**
Diagnosis	**ER+ HER2- breast cancer**	Accession ID	**V010900AA001-1**	Phone	**(833) 537-1819**
Medical Record #	**ID400231**	Report Date	**08/15/2026**	Fax	**(833) 874-0918**
Internal Patient ID	**10000090**	Test Number	**1**		

Northstar Select® Results

2 **informative genomic alterations identified**

Summary of Informative Genomic Alterations

Detected Genomic Findings	Associated FDA-Approved and/or Guideline-Recommended (*G*) Therapies			Clinical Trials	VAF / Copy number
	✓ **Approved** in indication	✓ **Approved** in other indication	✗ Associated with **resistance**		
PIK3CA N345K	• **alpelisib/fulvestrant**^{G} • **capivasertib/fulvestrant** • **inavolisib/palbociclib/ fulvestrant**			**10**	0.92%
PIK3CA P539R	• **alpelisib/fulvestrant**^{G} • **capivasertib/fulvestrant**^{G} • **inavolisib/palbociclib/ fulvestrant**^{G}			**10**	0.68%

Microsatellite Instability-High

Microsatellite Instability-High (MSI-H) is reported here as detected/not detected based upon mutation analysis at curated genome-wide MSI sites and subsequent bioinformatic algorithm calculation.

Not Detected Detected

Summary of Guideline-Recommended Genes Evaluated

The following guideline-recommended genes for Breast Cancer were evaluated. Variants not listed in the tables above are considered 'Not Detected'. For a complete list of genes tested, refer to the Methods section of the report.

AKT1 *BRAF* *BRCA1* *BRCA2* *ERBB2 (HER2)* *ESR1* *FGFR1* *FGFR2* *FGFR3* *NTRK* *PALB2* *PIK3CA* *PTEN* *RET*

Patient Name	**Jane Jones**	Date of Birth (Age)	**11/27/1940 (85 yrs)**	Assigned Sex at Birth	**Female**
Diagnosis	**ER+ HER2- breast cancer**	Internal Patient ID	**10000090**	Report Date	**08/15/2026**

Additional Genomic Findings

Genes with copy number signal indicating potential for aneuploidy

Copy number signal indicating potential aneuploidy was not detected in any gene.

Changes in copy number due to biological factors including aneuploidy can confound the accurate detection of clinically informative copy number alterations (CNAs). The following genes have deviations in copy number signal but are not called for copy number amplification/loss due to observed patterns of copy number signal (i.e. aneuploidy) within the chromosomal arm and/or other chromosomal arms of the sample.

Variants of Unknown Significance (VUS)

EGFR c.1831G>A (p.A611T), 0.66%
NOTCH1 c.4771C>T (p.H1591Y), 49.87%

The clinical relevance of these variants is currently unknown. Therefore, the functional impact of targeting these variants cannot be determined at this time.

Clinical Trial Availability

Clinical trial matches are displayed based upon somatic variant detection and patient demographic and diagnostic information provided on the test requisition form. Trials are matched within 500 miles of the ordering provider's location. This list is neither comprehensive nor a guarantee of eligibility, as many other requirements must be met prior to enrollment.

For further details on eligibility, please visit **www.clinicaltrials.gov** and enter the NCT#.

Phase 3 NCT06982521	A Phase 3 Open-Label Randomized Study Assessing the Efficacy and Safety of RLY-2608 + Fulvestrant Versus Capivasertib + Fulvestrant as Treatment for PIK3CA-mutant Hormone Receptor Positive, Human Epidermal Growth Factor Receptor 2 Negative (HR+/HER2-) Locally Advanced or Metastatic Breast Cancer Following Recurrence or Progression On or After Treatment With a CDK4/6 Inhibitor **PIK3CA N345K** **PIK3CA P539R**	**Palo Alto, California (94304)** Stanford Women's Cancer Center
Phase 3 NCT06790693	A Phase III, Multicenter, Randomized, Double-Blind, Placebo-Controlled Study Evaluating the Efficacy and Safety of Inavolisib Plus a CDK4/6 Inhibitor and Letrozole Versus Placebo Plus a CDK4/6 Inhibitor and Letrozole in Patients With Endocrine-Sensitive PIK3CA-Mutated, Hormone Receptor-Positive, HER2-Negative Advanced Breast Cancer **PIK3CA N345K** **PIK3CA P539R**	**Palo Alto, California (94301)** Palo Alto Medical Foundation Research Center
Phase 3 NCT07174336	A Phase 2, Randomized, Open-Label Dose Optimization and Phase 3, Randomized, Double-Blind, Placebo-Controlled Study of LY4064809 Combined With a CDK4/6 Inhibitor and Endocrine Therapy in Adults With HR+, HER2-Advanced Breast Cancer With a PIK3CA Mutation Who Received No Prior Treatment for Advanced Breast Cancer (PIKALO-2) **PIK3CA N345K** **PIK3CA P539R**	**Greenbrae, California (94904)** Marin Cancer Care
Phase 2 NCT07368998	A Phase II, Randomized, Open-Label Study Evaluating Two Inavolisib Dose Levels in Combination With Fulvestrant in Participants With PIK3CA-Mutated, HR-Positive, HER2-Negative Locally Advanced or Metastatic Breast Cancer **PIK3CA N345K** **PIK3CA P539R**	**Los Angeles, California (90017)** Los Angeles Cancer Network
Phase 2 NCT07054190	A Phase II, Open-Label, Multicenter Study Evaluating the Safety and Efficacy of Neoadjuvant Treatment With Inavolisib Combinations in Patients With Untreated, Early-stage, PIK3CA-Mutated Breast Cancer **PIK3CA N345K** **PIK3CA P539R**	**Los Angeles, California (90017)** Los Angeles Cancer Network
Phase 1/Phase 2 NCT04802759	A Phase Ib/II, Open-Label, Multicenter, Randomized Umbrella Study Evaluating the Efficacy and Safety of Multiple Treatment Combinations in Patients With Breast Cancer (MORPHEUS-BREAST CANCER) **PIK3CA N345K** **PIK3CA P539R**	**Stanford, California (94305)** Stanford Cancer Institute (SCI)

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Phase 1/Phase 2 NCT06993844	A Phase 1/2, Open-label, First-in-human Study of the Safety, Tolerability, Pharmacokinetics, and Preliminary Efficacy of ETX-636, a Pan-mutant-selective PI3Ka Inhibitor, as Monotherapy and in Combination With Other Anticancer Therapies in Participants With Advanced Solid Tumors **PIK3CA N345K** **PIK3CA P539R**	**San Francisco, California (94158)** UCSF Helen Diller Family Comprehensive Cancer Center
Phase 1/Phase 2 NCT05455619	Study to Assess the Safety and Efficacy of Evexomostat (SDX-7320) in Combination With A PI3K or Akt Inhibitor Plus Fulvestrant in Patients With HR+, HER2-, Metastatic Breast Cancer With PI3K Pathway Alteration(s) **PIK3CA N345K** **PIK3CA P539R**	**Newport, California (92663)** Hoag Memorial Hospital Presbyterian
Phase 1/Phase 2 NCT07287917	A Phase 1b/2 Trial Investigating the Safety and Efficacy of Oral AMXT 1501 and Oral DFMO in Combination With Standard of Care in Patients With Advanced Solid Tumors Who Progressed After Prior Therapies **PIK3CA N345K** **PIK3CA P539R**	**Los Angeles, California (90025)** START Los Angeles
Phase 1/Phase 2 NCT06736704	A Phase 1/2, Open-Label Dose Escalation and Expansion Study of SNV4818 as Monotherapy or in Combination With Other Anticancer Agents in Participants With Advanced Solid Tumors **PIK3CA N345K** **PIK3CA P539R**	**Los Angeles, California (90025)** The Angeles Clinic and Research Institute, A Cedars-Sinai Affiliate

Variant Details

PIK3CA

PIK3CA is an oncogene that encodes phosphatidylinositol 4,5-bisphosphate 3-kinase catalytic subunit alpha isoform (p110alpha), a lipid kinase that converts PIP2 to PIP3 at the plasma membrane, activating the AKT/mTOR signaling axis to promote cell growth, survival, and proliferation [1]. In cancer, gain-of-function variants constitutively activate p110alpha and amplifications increase p110alpha expression, driving persistent AKT/mTOR signaling and tumor cell proliferation and survival [2, 3].

PIK3CA N345K

Gene	**PIK3CA**
Nucleotide	**NM_006218.4: c.1035T>A**
Amino Acid	**p.N345K**
Exon	**5**
Biomarker Type	**activating**

PIK3CA N345 is located in the C2 domain of p110-alpha, and it has been shown by crystallography to interact with the PI3K regulatory subunit, p85 [4]. Cell-based assays demonstrate that the N345K mutation leads to reduced inhibition by p85, increased activity of PI3K, cellular transformation, and sensitivity to alpelisib [5, 6, 7, 8, 9, 10]. PIK3CA N345K has been reported in an ER positive breast cancer case who developed fulvestrant resistance; N345K was reported to decrease fulvestrant sensitivity in a breast cancer cell model [5].

PIK3CA P539R

Gene	**PIK3CA**
Nucleotide	**NM_006218.4: c.1616C>G**
Amino Acid	**p.P539R**
Exon	**10**
Biomarker Type	**activating**

PIK3CA P539R is a missense alteration that occurs within the helical domain of the p110-alpha protein (Uniprot). This alteration has been reported to result in increased cell growth, as well as kinase activity and cellular transformation in preclinical assays [6, 2].

Interpretation

Genomic alterations (SNVs/indels/fusions) were detected in the cell-free DNA (cfDNA) isolated from the patient's blood specimen. The variant frequency of the mutations, reported as variant allele fraction (VAF), is calculated and reported for detected SNVs and indels. These alterations may be informative to cancer treatment response and/or clinical trial eligibility. While somatic actionability is of primary concern when determining report inclusion, some variants may be included without matched actionable outcomes. These variants may be critical to carcinogenesis in the patient's tumor.

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Diagnosis	**ER+ HER2- breast cancer**	Internal Patient ID	**10000090**	Report Date	**08/15/2026**

Methods and Limitations

Northstar Select® is a next generation sequencing (NGS)-based in vitro diagnostic test for detection of substitutions (SNVs), insertion and deletion alterations (indels), selected genes' copy number alterations (CNAs, including amplifications and deletions) and selected gene-rearrangements in a total of 102 genes (Table 1), as well as microsatellite instability-high (MSI-H) status. Cell-free DNA (cfDNA) is extracted from plasma, and the targeted regions, encompassing >380kb, are amplified and sequenced. The sensitivity of the test in an accuracy study is 100% (95% CI: [99.84%, 100.00%]) for SNVs and indels for variant allele fraction (VAF) ≥ 0.5%, 100% (95% CI: [96.7%, 100%]) for fusions examined in the assay analytical validation, and 100% (95% CI: [76.84%, 100.00%]) for CNAs. The limit of detection of the assay (sensitivity ≥ 95%) is 0.15% VAF for SNVs and small indels (<40bp) and 0.30% for fusions. However, key actionable variants may be reported at lower VAFs where technically feasible. The overall base-wise specificity of the Northstar Select test is >99.99%, leading to high confidence in true positive reporting for variants with VAF >0.2%, despite the expansive genomic loci targeted and assayed. For CNAs, the LOD for gene amplifications is 2.11 copies and for loss is 1.8 copies. CNA LOD is subject to biological noise, with less aneuploid samples having a superior LOD. The copy number displayed on the report represents gene copies in liquid. Thresholds have been set to report homozygous loss for deletions, focal amplifications, or high-level amplifications (e.g. >6 copies estimated in the solid tumor). Observed increased or decreased copy number may not be called as CNAs despite potentially reflecting a characteristic (i.e., aneuploidy) of the tumor biology. These genes with copy number signal indicating potential for aneuploidy are listed in the corresponding section of the report (except for the AR gene located on the X-chromosome, which will not be subject to this additional analysis). MSI score is calculated using a count of somatic indel mutations in targeted microsatellite sites; based upon meeting threshold, MSI-H status is reported when detected. The sensitivity of the MSI component is 100% (95% CI: [97.72%, 100.00%]) for MSI-H at a tumor fraction of ≥ 0.5%, and the limit of detection (sensitivity ≥ 95%) of the assay for MSI-H is 0.07% tumor fraction. An MSI-Indeterminate result is an inconclusive result as it does not suggest the presence or absence of MSI-H in the patient. For certain cfDNA sample or variant characteristics, such as low cfDNA input or high level of cancer-associated chromosomal copy abnormality, the analytical sensitivity may be reduced.

Variants detected in the cfDNA are aligned to the hg19 reference genome. Informative genomic alterations are potentially actionable or biologically relevant variants based on evidence from medical and scientific literature. Variants of unknown significance (VUS) are genomic alterations that do not have sufficient evidence to determine biological/clinical significance. Variants classified as benign, likely benign, and variants likely originating from a pseudogene are not reported.

Genomic profiling of tumors can detect alterations not associated with the tumor itself but due to clonal hematopoiesis (CH). This assay cannot differentiate whether variants detected in the cfDNA are derived from a patient's solid tumor or clonal blood cells. Variants indicating CH, which are found most commonly in specific genes, may be detected, especially in older individuals or previously systemically treated patients (e.g., chemotherapy), and may impact targeted treatment efficacy. Clinical guidelines state that caution is warranted in interpreting ctDNA-only somatic alterations in genes indicated for PARP inhibitors and actionable tumor suppressor genes [11]. In alignment with guideline recommendations, detected alterations in these genes and other genes that are most CH-prone are flagged: *ATM, ATR, BARD1, BRCA1, BRCA2, BRIP1, CDK12, CHEK1, CHEK2, FANCA, FANCL, JAK2, MLH1, MRE11, NBN, NF1, PALB2, PTEN, RAD51B, RAD51C, RAD51D, RAD54L,* and *SF3B1* [12, 13, 14, 11]. Buffy coat enriched for white blood cells is currently stored by the laboratory in case further testing is necessary. This report should not be used in place of a dedicated hematological evaluation. Any interpretations should take into consideration the patient's clinical context.

Reported treatments and trials should be comprehensively evaluated by the medical provider, as inclusion in the report is neither a guarantee of treatment/trial match, nor intended to be fully comprehensive. Resistance mutation treatment associations are reported according to documentation from the FDA and/or professional guidelines. Treatments and trials are reported based upon the information provided at the time of test requisition, including diagnosis, sex, age, and location, and may not account for all elements of the patient's medical history. Certain treatments may have specifications surrounding other clinical factors, such as HR/HER2-status or germline mutation origin. The ordering provider should consult official FDA-approval information for full approval specifications, as well as professional guidelines for complete recommendations. Inclusion in Northstar Select does not guarantee that reported treatments or trials will have an impact on clinical outcome.

This assay is validated for the detection of somatic variants, but was not developed to distinguish germline variants, which may predispose the patient to certain types of cancer, from detected somatic variants. Incidental findings will be interpreted and reported in the context of somatic disease. For variants with a VAF reported at >40%, separate germline testing may be recommended to identify and characterize such variants that may have hereditary implications.

Table 1: Genes on the Northstar Select panel Northstar Select reports single nucleotide variants, insertion and deletion variants (indels), and splice site mutations for clinically relevant exons in 99 genes, copy number amplifications in 20 genes, copy number loss in 7 genes, and fusion events in 10 genes, as detailed in the table below. This assay was designed for blood-based molecular profiling of solid tumors; while indicated for use, gene coverage may not meet guideline recommendations for certain cancer types such as sarcomas and lymphomas. This test does not guarantee nor exclude possibility of detection of additional alterations; in certain clinical scenarios, additional testing may be indicated.

AKT1, *AKT2*, *ALK*$^{\wedge}$, *APC*, *AR*$^{+}$, *ARAF*, *ARID1A*, *ATM*$^{-}$, *ATR*, *BARD1*, *BRAF*$^{\wedge+}$, *BRCA1*$^{-}$, *BRCA2*$^{-}$, *BRIP1*, *CCND1*$^{+}$, *CCND2*, *CCNE1*$^{+}$, *CD274*$^{+}$, *CDH1*, *CDK12*, *CDK4*$^{+}$, *CDK6*$^{+}$, *CDKN2A*$^{-}$, *CDKN2B*, *CHEK1*, *CHEK2*, *CTNNB1*, *DDR2*, *EGFR*$^{+}$, *ERBB2*$^{+}$, *ESR1*$^{+}$, *EZH2*, *FANCA*, *FANCL*, *FBXW7*, *FGFR1*$^{+}$, *FGFR2*$^{\wedge+}$, *FGFR3*$^{\wedge}$, *FGFR4*, *GATA3*, *GNA11*, *GNAQ*, *GNAS*, *H3-3A*, *H3C2*, *H3C3*, *HRAS*, *IDH1*, *IDH2*, *JAK2*, *JAK3*, *KEAP1*, *KIT*$^{+}$, *KRAS*$^{+}$, *MAP2K1*, *MAP2K2*, *MET*$^{+}$, *MLH1*, *MPL*, *MRE11*, *MSH2*, *MSH6*, *MTAP*$^{-}$, *MTOR*, *MYC*$^{+}$, *NBN*, *NF1*, *NOTCH1*, *NPM1*, *NRAS*, *NRG1*$^{\wedge\wedge}$, *NTRK1*$^{\wedge}$, *NTRK2*$^{\wedge\wedge}$, *NTRK3*$^{\wedge\wedge}$, *PALB2*, *PDGFRA*$^{+}$, *PIK3CA*$^{+}$, *PIK3R1*, *PMS2*, *POLD1*, *POLE*, *PTEN*$^{-}$, *PTPN11*, *RAD51B*, *RAD51C*, *RAD51D*, *RAD54L*, *RAF1*$^{+}$, *RB1*$^{-}$, *RET*$^{\wedge+}$, *RHOA*, *RIT1*, *ROS1*$^{\wedge}$, *SF3B1*, *SMAD4*, *SMO*, *STK11*, *TERT*, *TP53*, *TSC1*, *TSC2*, *VHL*

$^{\wedge}$ Northstar Select also reports fusion events for this gene
$^{\wedge\wedge}$ Northstar Select reports only fusion events for these genes, no SNVs and indels will be reported; *NTRK3* is limited to *NTRK3-ETV6* fusions only
$^{+}$ Northstar Select also reports copy number amplifications of this gene
$^{-}$ Northstar Select also reports copy number loss of this gene

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This NGS-based assay was developed and its performance characteristics determined by BillionToOne, Inc. It has not been cleared or approved by the U.S. Food and Drug Administration. BillionToOne, Inc. is regulated under CLIA. This test is used for clinical purposes. It should not be regarded as investigational or for research. This test was performed using BillionToOne's patented technology (www.billiontoone.com/patents).

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