nanoString

nCounter[®] Breast Cancer 360[™] Panel

Gene Expression Panel

Subtyping • Disease Progression • Tumor Biology • Signature Development

The nCounter Breast Cancer 360 panel and data analysis service provides a unique 360 degree view of gene expression for the breast tumor, microenvironment and immune response. Now researchers can more quickly decode the complexities of breast cancer biology, develop novel breast cancer gene signatures, and categorize disease heterogeneity using 48 biological signatures including signatures based upon the analytically validated PAM50 and Tumor Inflammation Signature (TIS) assays.^{12,3}



Product Highlights

- Expertly curated, comprehensive content includes 776 human genes across 23 key breast cancer pathways and processes
- Provides a unique 360 degree view of gene expression for the breast tumor microenvironment and immune response
- Interactive Breast Cancer 360 data analysis report available to expedite analysis to insight
 - 48 signatures across 13 categories measuring biological variables crucial to breast cancer tumor biology
 - Access to validated signatures: PAM50, Tumor Inflammation Signature (TIS), Risk of Recurrence (ROR)/Genomic Risk
 - Expanded evaluation of breast cancer subtypes including: PAM50 Signature, TNBC, and Claudin- Low Signature
 - Publication ready figures and statistical methods included for speed to presentation

Feature	Specifications
Number of Targets	776 (Human), Including internal reference genes
Sample Input - Standard (No amplification required)	50-300 ng
Breast Cancer 360 Panel Standard	Synthetic oligonucleotide pool corresponding to all panel gene targets used for normalization
Sample Type(s)	FFPE-derived RNA, total RNA, and cell lysates
Customizable	Add up to 55 unique genes with Panel Plus
Time to Results	Approximately 24 hours
Data Analysis	nSolver™ Analysis software, the ROSALIND® Platform, and the Breast Cancer 360 Data Analysis Report

Comprehensive Content for Evaluating Breast Cancer Biology

Content included in the Breast Cancer 360 panel allows for a comprehensive measurement of biological variables crucial to tumor progression and response to a wide-range of treatments. Both analytically validated and research signatures are enriched with potentially predictive gene involved in proliferation, endothelial, angiogenesis, cytotoxicity, stroma, inflammatory chemokines, and apoptosis.

There are 48 signatures featured in this panel, including two analytically validated signatures (PAM50 and Tumor Inflammation Signature), three breast cancer subtyping signatures, eight breast cancer biology-focused signatures, four breast cancer receptor signatures, 28 novel signatures measuring important tumor and immune activities adapted to decode breast cancer heterogeneity.

PAM50 Signature

This 50-gene signature measures a gene expression profile that allows for the classification of breast cancer into four biologically distinct subtypes and a prognostic score:

- PAM50 Subtype (Luminal A, Luminal B, HER2-Enriched, and Basal-like)
- Prosigna Score / Risk of Recurrence



Tumor Inflammation Signature

This 18-gene signature measures activity known to be associated with response to PD-1/PD-L1 inhibitors pathway blockade³.

- Includes 4 Areas of immune biology used to determine peripherally suppressed immune response and the identification of "hot" or "cold" tumors: Antigen Presenting Cells, T Cell/NK presence, IFNγ Biology, and T Cell Exhaustion
- Tissue-of-origin agnostic (Pan-cancer)
- Potential surrogate for PD-L1 and mutational load in research setting⁴



Breast Cancer 360[™] Signatures

Included in this panel are 48 signatures across 13 categories of breast cancer tumor biology to support the evaluation of pathways and processes, as well as novel signature development.

Breast Cancer Prognosis	Risk of Recurrence (ROR)/Genomic Risk*							
Breast Cancer Subtyping	PAM50 Molecular Subtypes*	Luminal A Correlation Value (PAM50)	Luminal B Correlation Value (PAM50)	HER2-Enriched Correlation Value (PAM50)	Basal-like Correlation Value (PAM50)	Claudin-Low Subtype Score	Triple Negative Subtype	
Breast Cancer Receptors	ESR1 Gene Expression	PGR Gene Expression	ERBB2 Gene Expression	AR Gene Expression				
Breast Cancer Signaling Pathways	ER Signaling	PTEN Gene Expression	CDK4 Expression	CDK6 Expression				
Tumor Mutational Response	HRD	BRCA	p53					
Tumor Regulation	Proliferation (PAM50)	Apoptosis	Differentiation	FOXA1 Gene Expression	Cell Adhesion	Mammary Stemness	RB1 Gene Expression	SOX2 Gene Expression
Tumor Immunogenicity	APM (Antigen Processing Machinery)							
Stromal Factors	Endothelial Cells	Stromal Abundance						
Inhibitory Metabolism	Нурохіа							
Inhibitory Immune Mechanisms	IDO1 Expression	PD-L1 Gene Expression	В7-Н3	TGF-Beta				
Anti-Tumor Immune Activity	Tumor Inflammation Signature (TIS)*	Interferon Gamma Signaling	MHC Class II Antigen Presentation	Cytotoxicity				
Inhibitory Immune Signaling	Inflammatory Chemokines	TIGIT Gene Expression	PD-L2 Gene Expression	PD-1 Gene Expression				
Immune Cell Abundance	Cytoxic Cell	CD-8+ T Cell	Macrophage	Mast Cell	Treg			

*Validated Signatures

Breast Cancer 360 Data Analysis Report

A detailed signature analysis report for the Breast Cancer 360 panel is available. Interactive reports and individual consultation provide information on all 48 signatures and 776 genes included in the panel.

- Customized, interactive reports prepared by NanoString's expert scientists and biostatisticians.
- PAM50 intrinsic subtype provided for each sample to determine luminal A, luminal B, HER2-enriched, or basal-like subtypes. The correlation value to each subtype is also provided for samples run on BC 360.
- Tumor Inflammation Score provided for each sample to determine "hot" and "cold" tumors.
- All your data undergoes QC and normalization (up front).
- Analysis includes sample signature score in relation to response, treatment and survival (if annotations available).
- Includes a one hour report out and consultation with an expert analyst.
- Time series analysis available to determine how signatures are changing through time.
- Publication ready figures and statistical methods included for speed to presentation

Molecular profiles have distinct gene expression



Signature scores are shown for the selected sample, with PAM50 subtype in the center. Scores range from approximately 0-10; for most scores, a value of 5 is average. Each unit increase in score corresponds to a doubling of the biological process it measures. Color denotes each signature's biological function.

Ordering Information

Product	Product Description	Quantity	Catalog Num- ber	
nCounter Human Breast Cancer 360 Panel	Gene Expression CodeSet profiling (776 genes) 756 breast cancer-related human genes + 20 internal reference controls. No Master Kit	12 Reactions	XT-CSO-BC360-2-12	
nCounter Human Breast Cancer 360 Panel Standard	Standard containing a pool of synthetic DNA oligonucleotides that correspond to the target sequence of each of the 776 unique probe targets in the panel	12 Reactions	PSTD-HBC360-2-12	
nCounter Analysis System Master Kit	Reagents, cartridges, and consumables necessary for sample processing on the nCounter Analysis System.	12 Reactions	NAA-AKIT-012	
nCounter SPRINT Cartridge 1 Cartridge, 12 Ianes	Sample Cartridge for nCounter SPRINT System	12 Reactions	SPRINT-CAR-1.0	
nCounter SPRINT Reagent Pack	nCounter SPRINT Reagent Pack containing Reagents A,B,C & Hybridization Buffer	192 Reactions	SPRINT-REAG-KIT	
Low Input RNA Reagent Kit	48rxn kit for profiling from low sample input amounts	48 Reactions	LOW-RNA-48	
nCounter Breast Cancer 360 Primer Pool	MTE primer pools for Low Input RNA profiling (776 genes) 752 breast cancer related human genes + 18 internal reference controls. Master Kit, RNA Low Input Kit & Panel CodeSet Required	12 Reactions	PP-HBC360-12	
Breast Cancer 360 Data Analysis Report	Data analysis report for Breast Cancer 360 Panel	Report purchased in 12 sample increments	Contact your local rep	

Selected Panel References

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- 2. Perou CM, Sørlie T, Eisen MB, et al. Molecular portraits of human breast tumours. Nature. 2000;406(6797):747-52.2.
- 3. Ayers, Mark, et al. "IFN-γ-related mRNA profile predicts clinical response to PD-1 blockade." The Journal of Clinical Investigation 127.8 (2017).
- 4. Haddad R., Abstract 6009, ASCO 2017
- 5. Prat A, Parker JS, Karginova O, et al. Phenotypic and molecular characterization of the claudin-low intrinsic subtype of breast cancer. Breast Cancer Res. 2010;12(5):R68.
- 6. Burstein MD, Tsimelzon A, Poage GM, et al. Comprehensive genomic analysis identifies novel subtypes and targets of triple-negative breast cancer. Clin Cancer Res. 2015;21(7):1688-98.
- 7. Troester MA, Herschkowitz JI, Oh DS, et al. Gene expression patterns associated with p53 status in breast cancer. BMC Cancer. 2006;6:276.
- Severson TM, Wolf DM, Yau C, et al. The BRCA1ness signature is associated significantly with response to PARP inhibitor treatment versus control in the I-SPY 2 randomized neoadjuvant setting. Breast Cancer Res. 2017;19(1):99.
- 9. Peng G, Chun-jen lin C, Mo W, et al. Genome-wide transcriptome profiling of homologous recombination DNA repair. Nat Commun. 2014;5:3361.

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