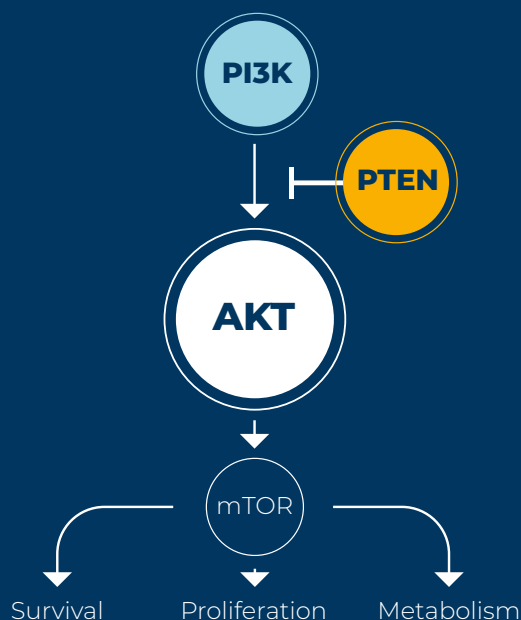


PIK3CA, AKT1, and PTEN biomarker testing in breast cancer

Testing patients' HR+/HER2- metastatic breast cancers for *PIK3CA*, *AKT1*, and *PTEN* alterations is recommended by NCCN Clinical Practice Guidelines in Oncology (NCCN Guidelines®).¹



1. The PI3K/AKT/PTEN pathway mediates critical cellular processes, such as cell proliferation, survival, and metabolism.²
2. Activation of the PI3K/AKT/PTEN pathway in HR+/HER2- breast cancer can occur through alterations in *PIK3CA*, *AKT1*, and/or *PTEN* genes.^{2,3}
3. Abnormal PI3K/AKT/PTEN pathway activation mediates resistance to multiple therapies in breast cancer, including ETs and CDK4/6is.^{1,4}

Adapted from Paplomata E, O'Regan R. *Ther Adv Med Oncol*. 2014;6(4):154-166.

PIK3CA, AKT1, and PTEN alterations in HR+ breast cancer



Patients only need to be positive for at least one *PIK3CA*, *AKT1* or *PTEN* alteration for their results to be actionable¹

NCCN makes no warranties of any kind whatsoever regarding their content, use or application and disclaims any responsibility for their application or use in any way.

©2024 AstraZeneca. All rights reserved.
US-88394 Last Updated 4/24

The importance of testing for *PIK3CA*, *AKT1*, and *PTEN* biomarkers with NGS



Whom to test

- Patients with HR+/HER2- locally advanced or metastatic breast cancer^{6,7}



When to test

- NCCN Guidelines[®] recommend ordering an NGS panel test during the initial patient work-up for metastatic disease^{1,6}
NGS panel tests can also be ordered after progression on first-line treatment⁶



How to test

- Utilize NGS^{*8}

The NGS testing process



Obtaining the sample⁸

Tissue biopsy: use archival primary or metastatic breast cancer tissue

- Surgically resected tumor tissue, or other biopsy samples (core needle, FNA), with sufficient cellularity can be used



Sample preparation⁸

Formalin-fixed paraffin-embedded specimen or fresh and/or frozen samples



Performing the test⁸

Send the NGS test to a commercial testing lab or perform NGS internally at your institution



Interpreting the results⁹

Biomarkers are classified according to their clinical significance:

- Actionable biomarker detected
- Potentially actionable biomarker detected
- No actionable biomarker detected



Clinical implications⁹

Results from the NGS report can inform on:

- Treatment options based on identified biomarker(s)
- Potential treatment resistance and disease progression
- Clinical trial eligibility

The NCCN Guidelines recommend NGS testing for detection of actionable biomarkers, such as *PIK3CA*, *AKT1*, and *PTEN*, in HR+/HER2- aBC^{†1}

NCCN makes no warranties of any kind whatsoever regarding their content, use or application and disclaims any responsibility for their application or use in any way.

*Although PCR can be used as a single gene test, NGS can provide more comprehensive information about the PI3K/AKT/PTEN pathway.

[†]aBC, advanced breast cancer, including locally advanced (inoperable) and metastatic breast cancer

NGS panel tests identify the most actionable biomarkers in breast cancer

The NCCN Guidelines recommend comprehensive germline and somatic profiling at metastatic breast cancer diagnosis during the initial work-up.¹ By ordering and performing NGS testing at metastatic diagnosis, the MDT can plan the patient's treatment throughout their breast cancer journey and be prepared for the next step.

National Comprehensive Cancer Network® (NCCN®) recommended biomarker testing for recurrent unresectable (local or regional) or stage IV (M1) disease¹

	Biomarkers	NGS	PCR	FISH/ISH	IHC	Germline Sequencing
Biomarkers associated with FDA-approved therapies ^{1,7,10}	<i>PIK3CA</i>	✓	✓			
	<i>AKT1</i>	✓				
	<i>PTEN</i>	✓				
	<i>ESR1</i>	✓	✓			
	<i>NTRK</i> fusion	✓	✓	✓		
	MSI-H/dMMR	✓	✓		✓	
	TMB-H	✓				
	<i>RET</i> -fusion	✓				
	PD-L1 ¹⁰				✓	
	HER2 (HER2 IHC 1+ or 2+/ISH negative)			✓	✓	
	Germline <i>BRCA1/2</i>					✓
Emerging biomarkers ¹	<i>HER2</i> activating mutations	✓				
	Somatic <i>BRCA1/2</i> mutations	✓				
	Germline <i>PALB2</i>					✓

Despite NCCN recommendations to order NGS testing, all eligible patients with HR+/HER2- mBC may not be undergoing NGS testing¹¹

Ensure all patients with HR+/HER2- mBC are tested with NGS at metastatic diagnosis or, at the latest, after progression on first line treatment



Look back to see if patients who have previously been tested with NGS have *PIK3CA*, *AKT1*, and/or *PTEN* alterations

Most common tissue-based NGS test panels include *PIK3CA*, *AKT1*, and *PTEN* genes

Send-out vs in-house	NGS Test Name	Number of genes	Sample requirements	PI3K/AKT/PTEN pathway genes included within panel
Send-out (centralized)	FoundationOne®CDx ^{9,12}	324	>20% tumor nuclei	<i>PIK3CA</i> , <i>AKT1</i> , and <i>PTEN</i> are detected
	NeoGenomics NeoTYPE® Breast Tumor Profile ^{13,14}	54		
	NeoGenomics Neo Comprehensive™ Solid Tumor ¹⁵	517		
	Tempus xT ¹⁶	648		
	Caris® Life Science Molecular Intelligence® Tumor Profiling ¹⁷	23,000+		
	Labcorp OmniSeq INSIGHT® ^{18,19}	523	>10% minimum tumor nuclei	
	Quest Diagnostics™ Solid Tumor Core Panel ²⁰	49	>20% tumor nuclei (>10% minimum)	
In-house (decentralized)	illumina® TruSight™ Oncology 500 ²¹	523	40 ng	
	Thermo Fisher OncoPrint™ Precision Assay GX ²²	50	>20% tumor nuclei	
	Thermo Fisher OncoPrint™ Focus Assay ²³	52	10 ng	<i>PIK3CA</i> and <i>AKT1</i> are detected

Speak to your multidisciplinary team to ensure *PIK3CA*, *AKT1*, and *PTEN* alteration status is identified for patients with HR+/HER2- advanced breast cancer—this could be discussed at the molecular tumor board.

Find out more at <https://www.azprecisionmed.com/tumor-type/breast-cancer/PIK3CA-AKT1-PTEN.html>

aBC, advanced breast cancer; AKT, serine/threonine protein kinase; *AKT1*, serine/threonine protein kinase 1; *BRCA1/2*, BRCA1/2, BRCA2, Breast Cancer susceptibility gene 1/2; CDK4/6, cyclin-dependent kinase 4/6 inhibitor; ctDNA, circulating tumor DNA; dMMR, deficient mismatch repair; *ESR1*, estrogen receptor 1; ET, endocrine therapy; FDA, US Food and Drug Administration; FISH, fluorescence in situ hybridization; FNA, fine-needle aspiration; HER2, human epidermal growth factor receptor 2; HR, hormone receptor; IHC, immunohistochemistry; ISH, in situ hybridization; mBC, metastatic breast cancer; MSI-H, microsatellite instability-high; mTOR, mammalian target of rapamycin; NCCN, National Comprehensive Cancer Network; NGS, next-generation sequencing; *NTRK*, neurotrophic tropomyosin receptor kinase; *PALB2*, partner and localizer of *BRCA1/2*; PCR, polymerase chain reaction; PD-L1, programmed death-ligand 1; PI3K, phosphoinositide 3-kinase; *PIK3CA*, phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit alpha; *PTEN*, phosphatase and tensin homolog; *RET*, rearranged during transfection; TMB-H, tumor mutational burden-high.

1. Referenced with permission from the NCCN Clinical Practice Guidelines in Oncology (NCCN Guidelines®) for Breast Cancer V.2.2024. © National Comprehensive Cancer Network, Inc. 2024. All rights reserved. Accessed March 26, 2024. To view the most recent and complete version of the guideline, go online to NCCN.org 2. Miricescu D, Totan A, Stanescu-Spinu II, et al. *Int J Mol Sci*. 2020;22(1):173. 3. Smyth LM, Zhou Q, Nguyen B, et al. *Cancer Discov*. 2020;10(4):526-535. 4. Papolomata E, O'Regan R. *Ther Adv Med Oncol*. 2014;6(4):154-166. 5. Martorana F, Motta G, Pavone G, et al. *Front Pharmacol*. 2021;12:662232. 6. Schwartzberg L, Kim ES, Liu D, et al. *Am Soc Clin Oncol Educ Book*. 2017;37:160-169. 7. Henry LN, Somerfield MR, Dayao Z, et al. *J Clin Oncol*. 2022;40(27):3205-3221. 8. Schmid S, Jochum W, Padberg B, et al. *ESMO Open*. 2022;7(5):100570. 9. Foundation Medicine. FoundationOne®CDx. Technical Information. https://www.accessdata.fda.gov/cdrh_docs/pdf17/P170019S006C.pdf. Accessed December 06, 2023. 10. Erber R, Hartmann A. *Breast Care (Basel)*. 2020;15(5):481-490. 11. Vanderwalde AM, Ma E, Yu E, et al. *J Clin Oncol*. 2021;39(suppl 28):288. 12. Foundation Medicine. FoundationOne®CDx. Specimen Instructions. https://www.foundationmedicine.com/sites/default/files/media/documents/2023-10/F1CDx_Specimen_Instructions%20%283%29.pdf. Accessed December 06, 2023. 13. Neo Genomics. NeoTYPE® Breast Tumor Profile Test Menu. <https://neogenomics.com/test-menu/neotype-breast-tumor-profile>. Accessed December 06, 2023. 14. Neo Genomics. NGS on Solid Tumor FFPE Tissue. https://neogenomics.com/sites/default/files/2024-01/Solid_Tumor_NGS_Specimen_Requirements_CORP-MRKT-0100_110823_Digital.pdf. Accessed April 16, 2024. 15. Neo Genomics. Neo Comprehensive – Solid Tumor. Test Menu. <https://neogenomics.com/test-menu/neo-comprehensive-solid-tumor>. Accessed December 19, 2023. 16. Tempus Oncology. Specimen Guidelines for Providers. https://www.tempus.com/wp-content/uploads/2022/09/Tempus-Onco_Specimen-Guidelines.pdf. Accessed December 06, 2023. 17. Caris Life Sciences®. Specimen Preparation Instructions. https://www.carislife.com/wp-content/uploads/2020/08/TN0252-v8_Specimen_Prep_Instructions_hi-rez.pdf. Accessed December 06, 2023. 18. OmniSeq INSIGHT®. Intended Use & Performance Specifications. <https://oncology.labcorp.com/sites/default/files/2022-04/OmniSeq-INSIGHT-INTENDED-US-PERFORMANCE-SPECS.pdf>. Accessed December 06, 2023. 19. OmniSeq INSIGHT®. Gene List. https://oncology.labcorp.com/sites/default/files/2023-03/OmniSeq_gene_list_DX_SS_L26488-1222-2.pdf. Accessed February 29, 2024. 20. Quest Diagnostics™ Solid Tumor Core Panel Test Detail. <https://testdirectory.questdiagnostics.com/test/test-detail/93234/solid-tumor-core-panel?q=93234&cc=MASTER>. Accessed December 06, 2023. 21. illumina®. TruSight™ Oncology 500 and TruSight Oncology 500 High-Throughput. <https://www.illumina.com/content/dam/illumina/gcs/assembled-assets/marketing-literature/trusight-oncology-500-data-sheet-m-gl-00173/trusight-oncology-500-and-ht-data-sheet-m-gl-00173.pdf>. Accessed December 06, 2023. 22. Thermo Fisher Scientific. OncoPrint™ Precision Assay. <https://assets.thermofisher.com/TFS-Assets/CSD/Flyers/oncomine-precision-assay-flyer.pdf>. Accessed November 17, 2023. 23. Thermo Fisher Scientific. OncoPrint™ Focus Assay. <https://www.thermofisher.com/uk/en/home/clinical/preclinical-companion-diagnostic-development/oncomine-oncology/oncomine-focus-assay.html>.