

Molecular In My Pocket™... ONCOLOGY: *Breast Cancer*

Tumor Group	Gene/Biomarker	Alterations	Indications	Result Interpretation/Significance	Assay Techniques
Invasive Ductal/Lobular Carcinoma	<i>BRCA1/2</i> **	Inactivating variants	Germline testing, therapeutic	Hereditary breast cancer, predicts response to PARP-inhibitors	CMA, MLPA, NGS, PCR, Sanger
	<i>CDH1</i> **	Inactivating variants	Germline testing, diagnostic	Inactivated in lobular carcinomas (in-situ and invasive), germline pathogenic variants associated with susceptibility to ILC and hereditary diffuse gastric cancer	CMA, IHC (E-cadherin), MLPA, NGS, PCR, Sanger
	<i>ERBB2</i>	Amplification, activating variants	Therapeutic	Molecular/intrinsic subtype classification, predicts response to anti-HER2 therapies	FISH, IHC, NGS
	ER/PR	Increased expression	Prognostic, therapeutic	Molecular/intrinsic subtype classification, predicts response to endocrine therapy	IHC
	<i>IDH2</i>	p.Arg172 hotspot variants	Diagnostic, possibly therapeutic in the future	Associated with tall cell carcinoma with reversed polarity	NGS, PCR, Sanger
	Ki67	Increased expression	Therapeutic	Predicts response to abemaciclib	IHC
	<i>PIK3CA</i>	Activating variants	Therapeutic	Predicts response to alpelisib; enriched in luminal A tumors	NGS, PCR, Sanger
	<i>PTEN</i> **	Inactivating variants	Germline testing	Cowden syndrome	CMA, MLPA, NGS, PCR, Sanger
	<i>STK11</i> **	Inactivating variants	Germline testing	Peutz-Jeghers syndrome	CMA, MLPA, NGS, PCR, Sanger
	<i>TP53</i> **	Inactivating variants	Germline testing	Li-Fraumeni syndrome, enriched in basal-like tumors	CMA, MLPA, NGS, PCR, Sanger
Salivary Gland-Type Neoplasms	<i>CRTC1::MAML2</i> <i>CRTC3::MAML2</i>	Fusion	Diagnostic	Associated with mucoepidermoid carcinoma	FISH, NGS, RT-PCR
	<i>ETV6::NTRK3</i>	Fusion	Diagnostic, therapeutic	Associated with secretory carcinoma, predicts response to larotrectinib and entrectinib	FISH, IHC (pan-TRK), NGS, RT-PCR
	<i>HMGA2</i> or <i>PLAG1</i>	Fusion	Diagnostic	Associated with pleomorphic adenoma	FISH, IHC (HMGA2, PLAG1), NGS, RT-PCR
	<i>HRAS</i>	p.Gly12, p.Gly13, p.Gln61 hotspot variants	Diagnostic, possibly therapeutic in the future	Associated with adenomyoepithelioma	IHC, NGS, PCR, Sanger
	<i>MYB::NFIB</i> and <i>MYBL1::NFIB</i>	Fusion	Diagnostic	Associated with adenoid cystic carcinoma	FISH, IHC/ISH (MYB), NGS, RT-PCR

Mesenchymal Neoplasms	<i>ALK</i>	Fusions	Diagnostic, therapeutic	Associated with inflammatory myofibroblastic tumor	IHC, FISH, NGS, RT-PCR
	<i>CTNNB1</i>	Activating variants	Diagnostic	Associated with desmoid fibromatosis	IHC, NGS, PCR, Sanger
	<i>EWSR1::DDIT3 and FUS::DDIT3</i>	Fusion	Diagnostic	Associated with myxoid round cell liposarcoma	FISH, NGS, RT-PCR
	<i>MDM2</i> or <i>CDK4</i>	Amplification	Diagnostic	Associated with well-differentiated liposarcoma and de-differentiated liposarcoma	FISH, NGS
	<i>MYC</i>	Amplification	Diagnostic	Associated with postradiation angiosarcoma	IHC, FISH, NGS
	<i>TERT</i>	Promoter variants	Diagnostic	Presence favors phyllodes tumor over fibroadenoma (both with <i>MED12</i> mutations)	NGS, PCR, Sanger
	<i>USP6</i>	Fusions	Diagnostic	Associated with nodular fasciitis	FISH, NGS, RT-PCR
	13q14	Deletion	Diagnostic	Associated with myofibroblastoma	FISH
Tumor-Agnostic Molecular Markers	Microsatellite instability (MSI)/mismatch repair (MMR) deficiency	Loss of MLH1, PMS2, MSH2, MSH6 expression and/or MSI-high status	Germline testing, therapeutic	Lynch syndrome, predicts response to immune checkpoint inhibitor (pembrolizumab; for unresectable/metastatic tumors)	IHC, PCR, NGS
	Tumor mutational burden (TMB)	SNVs and indels	Therapeutic	TMB ≥ 10 variants/Mb eligible for immune checkpoint inhibitor (pembrolizumab for unresectable/metastatic tumors)	NGS

*Legend: ** - germline risk factor; CMA - chromosomal microarray; FISH - fluorescence in situ hybridization; IHC - immunohistochemistry; indel - insertion-deletion; ISH - in-situ hybridization; LCIS - lobular carcinoma in situ; ILC - invasive lobular carcinoma; MLPA - multiplex ligation-dependent probe amplification; MMR - mismatch repair; MSI - microsatellite instability; NGS - next-generation sequencing; PARP - poly(ADP-ribose) polymerase; PCR - polymerase chain reaction; RT-PCR - reverse transcription polymerase chain reaction; SNV - single nucleotide variant*

Multi-Gene Expression Assays

Tumor Requirements	Assay Name	Assay Composition	Assay Results	Therapeutic Value	Prognostic Value
HR-positive, HER2-negative, 0-3 positive LNs, early stage IBC	Oncotype DX	16 cancer-related genes and 5 reference genes	Recurrence score (RS; 0-100)	Predictive for benefit of chemotherapy added to endocrine therapy	Prognostic for recurrence risk
	MammaPrint/Blueprint	70 genes (Mammaprint) and/or 80 genes (Blueprint)	MammaPrint index (MPI; -1.0 to +1.0) and low/high risk of recurrence; Blueprint molecular subtype result	Not determined	Prognostic for recurrence risk
	Prosigna	50 test genes and 8 housekeeping genes plus tumor size and nodal status	Risk of recurrence (ROR; 0-100)	Not determined	Prognostic for recurrence risk
	Breast Cancer Index	Ratio between <i>HOXB13:IL17BR</i> plus expression of 5 cell cycle-related genes	BCI Prognostic - low/high risk of recurrence; BCI Predictive - low/high likelihood of benefit	Predictive for extended endocrine therapy (beyond 5 years)	Prognostic for recurrence risk
	EndoPredict	8 target genes, 3 normalization genes, 1 control gene plus tumor size and nodal status	EPclin risk score (1.0-6.0), percent likelihood distant recurrence	Not determined	Prognostic for recurrence risk

Legend: HR - hormone receptor; IBC - invasive breast cancer; LN - lymph node

