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LONG COURSE ARTICLE

Prognostic and predictive parameters in breast pathology: a pathologist's primer

Kimberly H. Allison[1]

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Abstract

The pathologist's role in the breast cancer treatment team has evolved from rendering a diagnosis of breast cancer, to providing a growing list of prognostic and predictive parameters such that individualized treatment decisions can be made based on likelihood of benefit from additional treatments and potential benefit from specific therapies. In all stages, ER and HER2 status help segregate breast cancers into treatment groups with similar outcomes and treatment response rates, however, traditional pathologic parameters such as favorable histologic subtype, size, lymph node status, and Nottingham grade also have remained clinically relevant in early stage disease decision-making. This is especially true for the most common subtype of breast cancer; ER positive, HER2 negative disease. For this same group of breast cancers, an ever-expanding list of gene-expression panels also can provide prediction and prognostication about potential chemotherapy benefit beyond standard endocrine therapies, with the 21-gene Recurrence Score, currently the only prospectively validated predictive test for this purpose. In the more aggressive ER-negative cancer subtypes, response to neoadjuvant therapy and` the extent of tumor infiltrating lymphocytes (TILs) are more recently recognized powerful prognostic parameters, and clinical guidelines now offer additional treatment options for those high-risk patients with residual cancer after standard neoadjuvant therapy. In stage four disease, predictive tests like germline *BRCA* status, tumor *PIK3CA* mutation status (in ER+ metastatic disease) and PDL-1 status (in triple negative metastatic disease) are now used to determine additional new treatment options. The objective of this review is to describe the latest in prognostic and predictive parameters in breast cancer as they are relevant to standard pathology reporting and how they are used in breast cancer clinical treatment decisions.

Introduction

Every breast cancer pathology report includes multiple parameters that help inform prognosis and prediction in breast cancer. These parameters help clinical teams determine individualized treatment decisions for each breast cancer patient. The pathologist's role in cancer diagnosis has become that of a "Diagnostic Oncologist," with assessment of predictive pathology tests determining if a particular patient is eligible for specific therapies and prognostic parameters determining if additional treatments should be considered [1–6]. Pathologists should understand both the latest testing and reporting requirements and guidelines but also should have an understanding of their clinical relevance and be familiar with gray zones and unusual results so that they can serve as effective consultants.

While several of these parameters have had relevance for many decades, such as anatomic TNM staging, others have more recently proven their clinical relevance (especially in the metastatic setting) and still others are gaining new traction as evidence continues to evolve [7, 8]. The objective of this review is to describe the latest in prognostic and predictive parameters in breast cancer as they are relevant to standard pathology reporting and how they are used in breast cancer clinical treatment decisions. The reader will become familiar with how these parameters are used in the latest clinical guideline recommendations for prognostic/predictive testing in breast cancer and the evidence supporting their utility.

✉ Kimberly H. Allison
allisonk@stanford.edu

[1] Professor of Pathology, Director of Breast Pathology, Stanford University School of Medicine, Stanford, CA 94305, USA

Prognostic vs. predictive factors overview

Prognostic factors in breast cancer are those that define the natural history or outcomes of the disease, either without therapy or with standard therapy [9]. These factors help define who might need treatment in general based on the likelihood of a worse outcome. For example, estrogen receptor (ER) is a major prognostic factor in breast cancer because ER-negative breast cancers have a significantly worse 5-year overall survival than ER positive cancers [10, 11]. This major difference in outcomes also is supported by vast differences in the underlying biology between ER positive and ER-negative breast cancers [12–15]. Other traditional and evolving prognostic factors in breast cancer are listed in Table 1. Each factor plays a specific role in treatment pathways because of their association with systemic or local recurrence.

Predictive factors in breast cancer are those that are associated with likelihood of benefit from a specific treatment [9, 16]. These factors help inform which particular treatments may benefit an individual breast cancer patient. For example, ER is a prognostic factor because of its association with outcomes but it is also a predictive factor in breast cancer since cancers with ≥1% ER expression by immunohistochemistry testing receive a statistically significant disease free and overall survival benefit when treated with hormone-targeted therapies such a tamoxifen or selective estrogen receptor modulators (SERMS) [17, 18]. As shown in Table 1, the major predictive factors in early stage breast cancer remain biomarkers ER and HER2, but in stage four disease there has been a rapid evolution to include biomarkers predictive of drug benefit that include germline *BRCA* testing for all, and PDL1 immunoassays and *PI3KCA* mutation testing for specific subgroups of stage four patients.

Together, prognostic factors that help determine *who* needs treatment and predictive factors that help determine *what* specific treatments might be of benefit are used together to inform overall therapy choices for an individual patient. When multiple factors are combined together, overall treatment algorithms and calculators (such as the United Kingdom's *Predict* breast cancer tool) become more powerful at developing successful outcome predictors and treatment pathways for breast cancer patients (https://breast.predict.nhs.uk). The most powerful prognostic and predictive factors have become the major initial dividing points between different breast cancer treatment pathways in clinical guidelines such as the National Comprehensive Cancer Network (NCCN), American Society of Clinical Oncology (ASCO), and St Gallen International Consensus Guidelines. Probably the largest impact factors are ER and HER2 status, which used together create treatment-relevant subgroups of the four major breast cancer "intrinsic subtypes" (ER+/HER2−, ER+/HER2+, ER−/HER2+, and ER−/HER2−) within which different prognostic and predictive factors become relevant.

However, across all of these subtypes, the traditional pathologic TNM stage (based on size, lymph node status, and distant metastases) remains one of the most relevant prognostic factors. For each ER/HER2 subtype, there are significant differences in breast cancer specific survival by stage at diagnosis, with earlier detection making survival differences in each subtype [10, 19, 20]. For ER-negative cancers, differences in TNM anatomic stage remain relevant for the first 5 years of diagnosis, reflecting the more aggressive biology of these cancers and shorter timeline to recurrence and progression [10]. For ER positive cancers, differences in anatomic TNM stage at diagnosis remain relevant over many decades, reflecting the longer timeline to recurrence in this generally more indolent, slow-to-progress group of breast cancers [20].

Another factor that remains relevant across ER and HER2 subtypes is histologic subtype. In fact, the 2020 NCCN guidelines have histologic subtype as the very first branch point in treatment algorithms, even above ER and HER2 status [21] (www.nccn.org). While the ductal/not otherwise specified histologic group is a very broad diagnostic category without much prognostic relevance, cancers with a "favorable histologic type" have entirely different, less aggressive treatment pathways. Pure tubular, pure mucinous, pure cribriform, encapsulated papillary, or solid papillary are cancers that should be ER positive and HER2 negative, and in contrast to typical ER +/HER2− cancers, treatment beyond endocrine therapy is typically only considered in the lymph node positive setting for these special histologic subtypes because of their associations with excellent outcomes [22–28]. In addition, ER and HER negative good histologic subtypes such as adenoid cystic carcinomas and salivary or secretory carcinomas (or other rare histologic subtypes like low grade adeno-squamous or low grade fibromatosis-like carcinomas) are treated less aggressively than typical "triple negative" breast cancers with consideration for systemic therapies only in lymph node positive disease [23, 25, 29–35]. For these reasons, it is important for pathologists to follow strict diagnostic criteria when considering a favorable prognosis subtype diagnosis. If there are features that are atypical for a favorable histologic subtype diagnosis (such as high grade or HER2 positivity), these very specific diagnoses should be avoided and/or the pathologist should make it clear in their report that the outcomes typically associated with the standard or pure form of the favorable histologic subtype may not apply to a case with nonstandard features.

Table 1 Overview of prognostic and predictive pathology factors in nonmetastatic invasive breast cancer.

Factor	Prognostic	Predictive	Outcome associations	Clinical use/recommendation
Favorable histologic subtype (pure tubular, pure mucinous, adenoid cystic/salivary, secretory)	X		Good prognosis special histologic types are associated with more indolent behavior.	In NCCN guidelines, favorable histologic types are not considered for chemotherapy (regardless of size) unless lymph node positive. Pathologists should use strict criteria when diagnosing a favorable histologic subtype.
Nottingham score/grade	X		Higher grade cancers associated with more aggressive behavior, shorter times to recurrence/progression, more frequent response to neoadjuvant chemotherapy.	Used in combination with other factors in risk calculations, used in AJCC 8th ed. prognostic stage groups.
Size	X		Larger size at diagnosis (pT anatomic stage) associated with long-term risk of recurrence and breast cancer specific survival (especially in ER+ cancers).	Used in combination with other factors in risk calculations, used in AJCC 8th ed prognostic stage groups. Size thresholds (ex. 0.5 cm) are used in NCCN guidelines in determining when to consider chemotherapy ± targeted therapy (including when to consider 21-gene RS (Oncotype DX) testing). Size >5 cm is also considered an indication for postmastectomy radiation.
Lymph node status	X		Lymph node positive outcomes are substantially worse in all breast cancer subtypes.	Used in combination with other factors in risk calculations, used in AJCC 8th ed prognostic stage groups. In NCCN guidelines, lymph node positive cancers are considered for chemotherapy ± targeted therapies and radiation considerations are influenced (ex. including postmastectomy chest wall and/or axillary bed)
Margin status	X		Positive margins for invasion and 0.2 cm or closer margins for DCIS (without invasion) are associated with higher local recurrence.	Consensus guidelines recommend excision such that no tumor is at inked margins for invasive carcinoma (including associated DCIS) and in cases of DCIS without invasion, excision to 0.2 cm margins. Margin status also influences radiation postmastectomy considerations.
Associated DCIS	X		Extensive DCIS (or "extensive intraductal component") is associated with local recurrence when margins are close.	CAP Breast Cancer Reporting Protocol recommends reporting if "extensive intraductal component" because it may be considered in combination with margin status in radiation planning.
Lymphovascular invasion (LVI)	X		Extensive LVI associated with local recurrence and higher risk of lymph node positive disease.	Per NCCN, radiation post mastectomy should be considered if extensive LVI present (even when lymph node negative).
ER expression	X	X	ER-negative cancers have more aggressive behavior, shorter times to recurrence/progression, more frequent response to neoadjuvant chemotherapy.	NCCN, St Gallen and other treatment guidelines segregate overall treatment pathways by ER (and HER2) status.

Table 1 (continued)

Factor	Prognostic	Predictive	Outcome associations	Clinical use/recommendation
			ER positive (≥1%) by IHC is predictive of potential benefit from endocrine therapies.	Patients with ER positive cancers are considered for endocrine therapy regardless of size, grade, or lymph node status.
PgR expression	X		In ER positive cancers, the level of PgR expression is prognostic of overall outcomes, including on hormonal therapy (lower to negative PgR being associated with worse outcomes).	PR testing by IHC on invasive cancers can aid in the prognostic classification of cancers and serve as a control for possible false-negative ER results. Patients with ER-negative, PR-positive cancers may be considered for endocrine therapies, but the data on this group are noted to be limited. Used in some risk calculators.
HER2 status	X	X	HER2 positive cancers have more aggressive behavior, shorter times to recurrence/progression, more frequent response to neoadjuvant chemotherapy. HER2 positive status is predictive of potential benefit from HER2-targeted therapies (typically only when added to chemotherapies).	NCCN, St Gallen and other treatment guidelines segregate overall treatment pathways by HER2 (and ER) status. Patients with HER2 positive cancers are candidates for chemotherapy + HER2-targeted therapies (considered if ≤1 cm and standard if >1 cm or LN positive).
Proliferation (Ki67)	X		Higher Ki67 indices are associated with more aggressive behavior, shorter times to recurrence/progression, more frequent response to neoadjuvant chemotherapy. A decrease in Ki67 after neoadjuvant treatment is also associated with better outcomes.	Not currently used in most treatment guidelines. Used in some risk calculators.
Gene expression panels	X	X (21-gene recurrence score only)	A variety of gene-expression based panels exist with various prognostic and predictive abilities in specific settings.	The NCCN includes the 21-gene RS (Oncotype DX) in chemotherapy treatment decision-making in ER positive, LN negative cancers. Other settings and assays are currently considered prognostic only.
Neoadjuvant response (pCR vs. not or residual cancer burden)	X		In ER negative, HER2 positive cancers, better response to neoadjuvant therapy is strongly associated with improved survival (especially with a pCR). There is evolving evidence that residual cancer burden (RCB) is independently prognostic in all ER/HER2 subtypes at both 5 and 10 years.	If residual cancer is present after neoadjuvant chemotherapy, NCCN and St Gallen recommend considering additional treatment with capecitabine for triple negative cancers and switching to chemo-antibody drug, TDM-1, for HER2 positive cancers. Consider using standardized sampling and reporting as recommended by the RCB approach: see www3.mdanderson.org/app/medcalc
Tumor infiltrating lymphocytes	X		Higher TILs associated with better response to neoadjuvant treatments in triple negative and HER2 positive cancers. Higher TILs post treatment are also associated with better survival.	Not yet incorporated into standard treatment guidelines. If reported, use standardized approaches: see www.tilsinbreastcancer.org

Table 2 Predictive markers in breast cancer; thresholds and gray zones requiring confirmation, correlation, and explanation.

ER testing [38, 125]			
	Positive result	Gray zone/borderline result	Negative result
IHC nuclear staining:	>10%, strong intensity by IHC	1–10% or weak staining	<1% or 0%
	Correlation with histology recommended		
Confirmation:	No additional confirmation	Lab-specific SOP to confirm or adjudicate results	Lab-specific SOP to confirm or adjudicate results
Explanation/reporting:	Report positive results	Report as Low Positive with recommended comment about uncertainty of results close to thresholds	Report negative results, along with status of controls
HER2 testing [57, 58]			
	Positive result	Gray zone/borderline result	Negative result
IHC membranous staining:	3+ by IHC	2+ by IHC	0–1+ by IHC
	Correlation with histology recommended		
Confirmation:	No additional confirmation	In situ hybridization (ISH) as reflex test	No additional confirmation
Explanation/reporting:	Report as positive	Equivocal, defer to ISH testing	Report as negative
In situ hybridization testing (ISH), dual probe:	Group 1 by ISH	Groups 2–4 by ISH	Group 5 by ISH
	Correlation with histology recommended		
Confirmation:	No additional confirmation	Review concurrent IHC from same sample. If 2+ by IHC second observer recounts ISH.	No additional confirmation
Explanation/reporting:	Report as positive	Report as overall HER2 positive or negative based on combined results of IHC and ISH (per guidelines). Use recommended reporting comments to clarify limited data on these groups.	Report as negative

Threshold setting and gray zones in predictive vs. prognostic biomarker use

With biomarkers such as ER and HER2 being the first major branch point in treatment pathways, it is important to note that thresholds for a positive result were created for their predictive value, not necessarily for overall treatment pathways decisions. Setting thresholds for biomarkers depends on what you are trying to prognosticate or predict [36, 37]. When predictive, the threshold set will also depend to some extent on the risk-benefit profile of the drug the test makes the patient a candidate for. For example, the risk profile of endocrine therapies, like tamoxifen, is quite low for its potential benefit, so the threshold for a positive result is set quite low (at 1% of cancer cells staining by IHC) to include as many patients as possible that might benefit from this low-risk drug category [38, 39]. With this very sensitive definition of ER positive, a 50–66% reduction in recurrences and 30–40% reduction in overall mortality is observed for patients with ER positive breast cancers treated with endocrine therapy [17]. Even for the lowest levels of ER expression (10–19 fmols by ligand binding assay, which correlates with 1–10% protein expression by IHC) the limited evidence available points to a one third reduction in recurrences with 5 years of tamoxifen, even though many of these cancers have other characteristics more similar to ER-negative cancers [17, 18, 40–51]. For HER2 testing, a positive result should predict potential benefit from HER2-targeted therapies, which are currently routinely given with chemotherapy [52–58]. The risk profile of chemotherapy with HER2-targeted therapies is higher (and more expensive) than endocrine therapy alone with a high risk with both false positive and false-negative results. Testing strategies that use IHC testing as an initial HER2 screening test, with reflex to the more complex in situ hybridization (ISH) testing for IHC equivocal results (or dual IHC and ISH testing) helps to ensure the accuracy of HER2 results.

It is important to acknowledge that there will also usually be a gray zone near the threshold set for a positive vs. negative result, around which there will be both more variability in test results and less clear clinical implications [59]. Both the ER and HER2 testing ASCO/CAP guidelines updates have focused on fine tuning guidelines for these more gray zone results, recommending correlation, confirmation, and explanation in reporting for certain scenarios (see Table 2). For ER testing, the 2020 ASCO/CAP Guidelines Update recognized that cases with <10% or weak staining may need additional steps to confirm the

results and recommended that labs set up their own standard operating procedures (such as review of controls, having a second pathologist confirm results, or use a digital-aided interpretation) to confirm or adjudicate these results [38]. In addition, the update also created the ER Low Positive reporting category for cases with 1–10% ER staining, which is to be reported with a comment that acknowledges the limitations of the data on cancers with Low Positive ER. Similarly, HER2 testing gray zones include 2+ results by IHC (which should be reflexed to ISH testing) as well as the unusual result groups by ISH (Groups 2–4 in the 2018 ASCO/CAP HER2 testing update) [56, 58]. Similar to the strategy for ER testing near the threshold for positive, for unusual HER2 ISH group testing additional steps are recommended in the evaluation and work-up of these cases prior to reporting a final overall HER2 status. Reporting comments are also recommended to clarify issues with these unusual HER2 ISH result categories so clinicians and patients can better understand that the HER2 status is non standard. The same holds true for commercial tests like Genomic Health's 21-gene Recurrence Score (RS), which is now prospectively validated to predict the potential additional benefit of adding chemotherapy to endocrine therapy in ER positive, lymph node negative cancers [60]. Results that are in the middle of the RS range have less clear implications, that differ by age group, than results at the ends of the RS result spectrum [60].

Prognostic stage groupings

When individual factors are combined together, they can become even more powerful predictors of outcomes. With this concept in mind, the 8th edition of the AJCC Cancer Staging Manual expanded their traditional anatomic TNM factors to include tumor grade, ER, HER, and Oncotype RS as part of a new "prognostic stage groupings" to further refine outcome prediction [7, 8, 61]. Initially based on data from the MD Anderson Bioscore and Risk Score using California Cancer Registry (CCR) and National Cancer Database databases, it has since been further validated to outperform the anatomic stage using over 50,000 breast cancer patients in the CCR [62–65]. Using the prognostic stage, which combines the TNM stage with these additional factors, ~40% of breast cancer patients were restaged into higher or lower prognostic stage groups. This staging system is definitely more complex, requiring reference to large tables with over 150 possible combinations of factors listed. Initially, some combinations were not listed, requiring updating of the original version. As the AJCC prognostic stage groupings and other methods of prognostication continue to evolve, perhaps they will incorporate new prognostic factors in more dynamic ways using risk calculator tools or more dynamic risk assessments at different timepoints in disease progression [66, 67]. Risk calculator tools and nomograms already exist but are not necessarily validated in multiple data sets. As mentioned above, the United Kingdom's PREDICT breast cancer online calculator tool (https://breast.predict.nhs.uk) is an example of one of these tools. It was created to using outcomes data from the National Health Service to incorporate standard clinicopathologic parameters like age, menopausal status, ER, HER2, Ki67, tumor size, grade, and lymph node status to calculate overall 5, 10, and 15 years survival differences with and without additional therapies [68–70].

ER and HER2 status have created groupings of breast cancers, within which, different factors are relevant. However, another important consideration is that different prognostic factors have very different relevance at different timepoints, from initial diagnosis where the initial question is whether a patient will need additional therapies beyond methods used for local control (surgery ± radiation), to post treatment when additional therapies may be needed if there is residual disease, to recurrence and progression. Traditionally, there has been a focus on initial diagnosis but a better understanding of indicators of outcomes in the post treatment and recurrence/progression timepoints has been a rapidly evolving area. Prognostic and predictive factors are presented in the next few sections grouped similar to treatment guidelines, with the various combinations of ER and HER2 status creating subgroups within the early stage vs. metastatic settings.

Prognostic and predictive factors in stage 1–3 ER positive, HER2 negative breast cancers

Because nonmetastatic ER positive, HER2 negative cancers are the largest group of breast cancers overall, much of the historical data on prognosis and outcomes were largely related to this group. Traditional factors like size, lymph node status, and age at diagnosis are highly relevant to recurrence risk in this group over many decades. Additional important prognostic segregators of outcome that reflect the spectrum of underlying biology of these cancers include Nottingham grade, proliferation, and PgR levels. Higher grade, more proliferative ER positive cancers have a higher risk of recurrence earlier than ER positive cancers with a low grade and proliferative rate. In addition, the high grade, high proliferation ER positive cancers potentially stand to benefit more from the addition of chemotherapy in addition to endocrine therapy. However, there are no specific predictive thresholds for grade or proliferation (such as scored with Ki67 IHC) that are clinically validated to predict

chemotherapy benefit, so these factors remain prognostic. In addition, since most ER positive cancers have very high levels of ER expression by IHC (rather than a spectrum of ER expression), the downstream hormone receptor, PgR, which has more of a spectrum of expression, turns out to be a more powerful prognostic indicator in ER positive cancers. ER positive breast cancers with low levels of PgR expression are believed to have a less "intact" or responsive hormone-driven pathway, and subsequently are hypothesized to have decreased responsiveness to endocrine therapy alone [71–77]. However, as with grade and proliferation rates, specific PgR IHC thresholds to predict decreased response to endocrine therapy or potential benefit from the addition of chemotherapy are not validated and therefore, PgR remains a prognostic rather than predictive factor.

What has emerged as a validated predictive factor to help determine potential chemotherapy benefit in the lymph node negative subset of ER positive, HER2 negative breast cancers is the 21-gene RS (Oncotype DX) [78, 79]. This RT-PCR assay generates a RS that is heavily weighted by the level of the five proliferation-related genes assayed but also includes PgR levels (as well as ER, HER2, and a few additional genes). The RS has now been prospectively validated in the TAILOR RX trial to predict the benefit of adding chemotherapy to endocrine therapy (a result which is modified by age) [79]. As such, it is the only validated predictive test in this setting. NCCN and ASCO guidelines currently recommend this test to predict which ER positive, HER2 negative, lymph node negative cancers >0.5 cm may benefit from the addition of chemotherapy [21, 80]. However, other treatment recommendations, such as the St Gallen consensus recommendations, recommend individualized decision-making based on multiple factors, recognizing that gene-expression profile testing may not be available in all settings [81, 82].

While predictive in lymph node negative cancers, it currently serves only as a prognostic test in lymph node positive, ER positive, HER2 negative patients, with the results of another prospective trial, the RxPONDER trial, anticipated to resolve if it is predictive or remains only prognostic in the lymph node positive population [83]. Other gene-expression tests have shown prognostic value as well, helping stratify which ER positive cancers are at a higher risk of recurrence, but have not shown specific predictive value to date [84–89].

Prognostic and predictive factors in stage 1–3 HER2 positive breast cancers

HER2 positivity is considered a poor prognostic factor in both ER positive and negative breast cancers. HER2 positive cancers are typically higher grade, with higher proliferative rates and are more rapidly progressive than ER positive, HER2 negative cancers. Outcomes for HER2 positive cancers were similar to or worse than triple negative breast cancers prior to the advent of HER2-targeted therapies. In addition to being prognostic, HER2 is a powerful predictive factor, used to select which patients might benefit from HER2-targeted therapies in combination with chemotherapy [90–95]. Because of its aggressive biology, chemotherapy plus HER2-targeted therapy with trastuzumab is considered in invasive HER2 positive cancers of any size or lymph node status, and higher risk features such as size ≥2 cm or positive lymph nodes are used to determine if a *second* HER2-targeted therapy (pertuzumab) can be added to additionally reduce risk [96–98]. De-escalation of the therapy regimen for early stage HER2 positive cancers is considered if the cancer is <1 cm and lymph node negative because of good outcomes in this setting. Unlike the ER positive, HER2 negative group, grade, and proliferation are not considered useful prognostic parameters in this group. Instead, response to neoadjuvant therapies, frequently measured as the "residual cancer burden" is a more powerful prognostic indicator, with excellent long-term outcomes for HER2 positive cancers treated to a complete pathologic response and those with a high residual disease burden at the highest risk of progression [99–108]. It is for this reason that clinical treatment guidelines consider adding on additional treatments like the antibody-drug conjugate (ADC), TDM-1 (ado-trastuzumab-emtansine), to patients that remain at high risk of recurrence because of residual HER2 positive cancer after neoadjuvant treatment [109]. This drug, first approved in the metastatic HER2 positive setting, delivers a powerful chemotherapy intracellularly once the trastuzumab HER2 antibody binds to HER2 positive cells and avoids many of the side effects of traditional systemic chemotherapy regimens. Other ADCs are being developed and tested that may target lower levels of HER2, which if approved, may dramatically alter how HER2 testing is used as a predictive test [110].

Tumor infiltrating lymphocytes (TILs) have also turned out to be prognostic in HER2 positive cancers. HER2 positive cancers with higher TIL levels are reported to have better response rates to neoadjuvant treatment, which typically correlates with overall survival rates as well [111–115]. The hypothesis is that more TILs present may create a "primed" immune environment, helping to create durable responses to eliminating a cancer. While TILs reporting has not uniformly become standard practice, there are guidelines for standard evaluation and reporting [111, 116–118] (www.tilsinbreastcancer.org).

Prognostic and predictive factors in stage 1–3 ER-negative, HER2 negative breast cancers

Similar to the HER2 positive cancers, ER-negative/HER2 negative cancers are typically higher grade and higher proliferation, making these factors less prognostic in this higher risk group. Most treatment guidelines will only risk not treating these aggressive cancers with chemotherapy when they are lymph node negative and less than 0.5 cm, with consideration for treatment when pN1mi or the primary is 0.6–1.0 cm. Chemotherapy is more standard for any lymph node positive triple negative breast cancer or one that is over 1 cm. The exception to this would be the unusual good prognosis triple negative histologic subtypes as mentioned above (adenoid cystic and some other salivary type carcinomas, etc.). Response to neoadjuvant chemotherapy is a powerful prognostic factor in the treated group of triple negative breast cancers (similar to HER2 positive cancers) with additional therapy considered if there is residual disease post treatment (with agents such as capecitabine, an oral chemotherapy) [80]. Similarly, TILs have shown prognostic relevant in determining better response to neoadjuvant chemotherapy [111, 112, 116–118]. Many triple negative breast cancers have high genomic instability and there is great interest in determining if immunogenic markers of this instability (such as BRCA mutations or Tumor Mutational Burden) might serve as a predictive biomarkers for additional therapies such as poly-ADP ribose polymerase (PARP) inhibitors and immunotherapies. However, these are still largely investigational in early stage triple negative disease, with approval for use currently only in the advanced or metastatic setting (see next section).

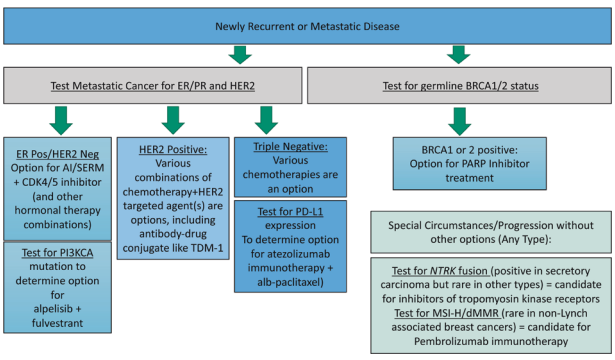

Fig. 1 Testing considerations for newly recurrent of metastatic breast cancer. All newly recurrent or metastatic breast cancer patients should have their disease sampled and tested for current ER/PR and HER2 status with additional testing dependent on results and possible treatment pathways as shown. All patients can also be considered for germline BRCA testing to determine if PARP inhibtor treatments are an option.

Predictive factors in stage 4 breast cancer

The testing and treatment landscape of metastatic breast cancer has changed dramatically in the last 5 years, with new predictive testing available to help guide treatment in this traditionally very challenging treatment group. (See Figure 1) In a more recent development, all patients with newly metastatic or recurrent breast cancers are now recommended to undergo germline BRCA1/2 testing since the identification of a significant germline mutation predicts potential benefit from adding PARP inhibitors (based on the OlympiAD and EMRACA trials) [119, 120]. In addition, any new metastatic breast cancer should be tested for ER and HER2, since the metastatic status can be different from the primary (although treatment path can also be based on the status of the primary cancer). Similar to in the nonmetastatic setting, ER and HER2 testing is predictive of benefit from specific therapies in the metastatic setting and results of these tests creates subgroups where additional tests may identify candidates for additional treatments.

Patients with ER positive, HER2 negative cancers may derive benefit from not only an aromatase inhibitor or SERM but also from the addition of a CDK4/6 inhibitor (such as abemaciclib, lapbociclib, or ribociclib). Based on results of the SOLAR1 trial, PIK3CA mutation testing (of the tumor or ctDNA in blood) may also identify patients who can benefit from an alpelisib–fluvesterant combination to inhibit the PI3K and ER pathways [121]. These oral medications can allow slowly progressive ER positive stage 4 breast cancer patients to save more aggressive chemotherapy regimens for later timepoints in progression or metastatic crisis.

Patients with ER-negative, HER2 negative metastatic disease now also have a predictive marker that is a companion diagnostic. These metastatic (or prior primary cancers) can be tested for PD-L1 expression using the SP142 IHC antibody and the Immune Cell scoring system to determine if immune therapy with atezolizumab with alb-paclitaxel chemotherapy may be of benefit [122]. Pembrolizumab (in combination with chemotherapy) was also recently shown to increase progression free survival in triple negative metastatic breast cancer in cancers that tested PD-L1 positive using the 22C3 antibody and a ≥ 10% threshold with the Combined Positive Score system. (KEYNOTE-355 unpublished data, ClinicalTrials.gov Identifier: NCT02819518) Pembrolizumab also may provide survival benefit for patients with MSI-H/dMMR positive cancers (agnostic of site of origin) and was also recently FDA-approved for treating tumor mutation burden high (≥10 mutations/megabase) metastatic cancers from any site [123] (KEYNOTE-158, NCT02628067). This testing and treatment landscape is rapidly changing.

Lastly, rare mutations like *NTRK* fusions, which are seen in over 90% of secretory carcinomas of the breast (and <5% of other breast cancers), allow for treatment in the metastatic setting with inhibitors of tropomyosin kinase receptors [124]. Frequently, comprehensive genomic profiling is performed on metastatic cancers in practice in an attempt to identify these targetable mutations.

Conclusions

Prognostic and predictive parameters in breast cancer have expanded as our understanding of their relevance in specific biologic subtypes and stages of breast cancer has deepened. New therapies, especially in the metastatic breast cancer setting, have resulting in new predictive tests. Clinical treatment guidelines continue to evolve with new data on these pathology parameters. Pathologists should remain up to date with current testing and treatment guidelines.

Compliance with ethical standards

Conflict of interest The author declares no conflict of interest.

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