Review Article

Genetic Panels in Breast Cancer: Current Guidelines

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Abstract

The breast is the most frequent site of cancer among women in both developed and in developing countries [1]. Breast cancer is the most frequent cause of cancer death in women from less developed countries and second most frequent (after lung cancer) among women in developed countries [1]. It is increasing in incidence in the developing world due to increased life expectancy, increased urbanization and the adoption of western lifestyles [2]. According to the World Health Organization (WHO), "Early detection in order to improve breast cancer outcome and survival remains the cornerstone of breast cancer control" [2]. Breast cancer staging also takes into account cancer's grade; the presence of tumor markers, such as receptors for estrogen, progesterone and HER2; and proliferation factors.Standard screening for new and recurrent breast cancer involves clinical breast exam and breast imaging. Given the recognized differences in breast cancer incidence, subtypes, and prognosis among women , it is important to evaluate potential biomarkers in the landscape of breast cancer subtypes ranging from DCIS to triple-negative breast cancer (TNBC) to determine if a simple blood test can enhance the diagnosis of this disease and knowing about stage of disease, and helping in proper treatment. In this review we tried to discuss role of genetic panels in breast cancer management.
Keywords: Breast cancer, WHO, Genetic Panels.

INTRODUCTION

Breast cancer is the second most common cancer worldwide after lung cancer, the fifth most common cause of cancer death, and the leading cause of cancer death in women [3]. Recent statistics suggest that about 1.3 million females develop breast cancer each year and about 465,000 of them succumb to the disease [4, 5]. Breast cancer can be diagnosed by Breast examination,mammogram,breast ultrasound,breast MRI,Biopsy. Although mammography screening is available, there is an ongoing interest in improved early detection and prognosis.

Biomarker analysis in cancer not only provides additional information about classical clinical factors, but also enables patients with a more favourable benefit–risk balance to receive certain treatments [3]. In breast cancer, biomarker analysis is routine practice. It originally began with testing for hormone receptor expression to guide tamoxifen therapy. The subsequent inclusion of targeted treatments against human epidermal growth factor receptor 2 (HER2) revolutionised the biomarker field. Molecular genomic testing provides clinicians with both prognostic (and sometimes predictive) information that can help individualize treatment and decrease the risk of over- or under-treatment.

Genomic Assays
Oncotype Dx

The test is valid for women with hormone sensitive breast cancer. It is most often used for women with early stage (DCIS, stages I and II node negative) disease. Results provide a statistical inference of chemotherapy benefit and likelihood of recurrence [6].

Oncotype Dx is a 21-gene (16 breast-cancer-related genes and 5 reference genes), reverse-transcriptase polymerase chain reaction (RT-PCR) assay .Oncotype DX uses FFPE from surgical specimens to categorize patients into one of three tiers based on a calculated Recurrence Score (RS) – low (<18), intermediate (18–30), and high (≥31–100) – reflecting their likelihood of distant recurrence in 10 years. In the Trial Assigning IndividuaLized Options for Treatment (Rx), also known as TAILORx, women with a Recurrence Score of less than 11 were found to have a <1% risk of recurrence in 10 years with receipt of endocrine therapy alone, further bolstering support for a paradigm shift away from mandatory chemotherapy within the context of multimodal

treatment. [7]. Results from the West German Study Group Phase III PlanB Trial provided additional, prospectively generated evidence that patients with an Oncotype Dx RS ≤11 could avoid chemotherapy without compromising outcomes, even if said patients had clinicopathologic characteristics that would otherwise point towards a high risk of recurrence [8]. Oncotype DX currently issues separate reports for LN− and LN+ (N1–3) patients. The RxPONDER Trial (Rx for Positive Node, Endocrine Responsive Breast Cancer) was initiated in 2011 to explore whether ER+, HER2− patients with limited nodal disease (1–3 LNs) and low to intermediate Oncotype DX scores would experience decreased survival if chemotherapy were omitted from their regimens; another aim of this trial is to determine whether there is an optimal RS cutoff point for these patients, above which chemotherapy should always be recommended [9]. The 21-gene Oncotype DX assay is mentioned in the NCCN guidelines as a possible consideration to help guide the addition of chemotherapy in patients with limited (1–3) positive nodes since there is ample data from the Southwest Oncology Group (SWOG) 8814 [10], the NSABP B-28 [11], and the studies mentioned above to suggest that it provides predictive utility of chemotherapy benefit in patients with limited nodal involvement. The results from the RxPONDER trial should help clarify the role of genomic testing with Oncotype DX in LN+ patients.However, it has been demonstrated to have a high false-negative rate for tumors that are HER2+ and therefore, is not indicated for use in HER2+ patients [12].

Mammaprint

MammaPrint®, which was first described in 2006, is a 70-gene DNA assay developed by Agendia (Irvine, CA), a commercial spin-off of the Netherlands Cancer Institute (NKI) and Antoni van Leeuwenhoek Hospital in Amsterdam [13]. It consists of a customized microarray slide that assesses in triplicate the mRNA expression of 70 genes initially identified in 78 tumors from a cohort of T1–2, LN-breast cancer patients under the age of 55 years at diagnosis and treated at NKI; 50% of these patients were ER+ [14]. The assay can use either fresh-frozen tumor samples or FFPE. The MammaPrint Index (i.e., score) ranges from −1 to +1; tumors with a Mamma Print Index of <0.4 are classified as having a low risk of distant metastasis in 10 years while those tumors with scores of ≥0.4 are at high risk for developing distant metastases in 10 years [15, 16]. MammaPrint was the first genomic assay approved by the FDA and is the most widely used breast cancer-specific genomic assay in Europe.

Mammaprint can be used to analyze both ER (−) and ER+ early stage (i.e. stage I or II) node negative (U.S. criteria; international criteria allow up to three positive nodes) invasive cancers [17].

Prosigna

The 50 gene assay, formerly called the PAM50 test, analyzes the activity of certain genes in node-negative (stage I or II) or node positive (stage II), hormone receptor-positive breast cancer patients. It provides individualized assessment of a patient's risk of recurrence at 10 years if given endocrine therapy alone [18].

Prosigna is based on a 50-gene RT-PCR microarray that uses its proprietary nCounter® digital technology to process postoperative FFPE samples of invasive carcinoma and assign tumors to one of four intrinsic subtypes: Luminal A, Luminal B, HER2+, and Basal-like. In addition, the Prosigna gene signature also generates an individualized Risk of Recurrence (ROR) score (high, intermediate, or low) representing an estimate of the likelihood of developing recurrent disease through an algorithm that takes into account intrinsic subtype, correlation between molecular subtype and a subset of proliferative genes, and tumor size on final pathology. It has been retrospectively validated in postmenopausal women receiving adjuvant endocrine therapy for both LN+ and LN− breast cancer and was cleared by the FDA for marketing as a prognostic tool in 2013 [19].

Breast Cancer Index

The Breast Cancer Index represents a combination of two diagnostic tests – the 2-gene, HoxB13/IL17BR ratio index (HI) and the Molecular Grade Index, a real-time RT-PCR, 5-gene microarray assay – that has been retrospectively validated to predict the likelihood of late (i.e., 5–10 years after treatment) recurrence as well as the likelihood of benefit from a 10-year course of adjuvant endocrine therapy in women with early-stage, LN−, ER+ breast cancer [20, 21]. Specimens can be FFPE or fresh frozen. It is not currently approved by the FDA for marketing in the US.

EndoPredict

The EndoPredict Test combines EndoPredict– an 8-gene, mRNA-based assay that uses RT-PCR on FFPE tumor samples – with patient tumor size and nodal status to assign patients with early-stage, ER+, HER2− breast cancer a score that reflects likelihood of distant recurrence within 10 years of diagnosis. Patients with a score of <3.3 are at low risk for recurrence, and those with a score of ≥3.3 are at high risk for recurrence [22]. The EndoPredict Test is not currently approved by the FDA for marketing in the US but is approved for use in Europe.

Genomic Grade Index

The Genomic Grade Index is a DNA microarray-based assay that uses FFPE tumor samples to measure the expression of 97 genes and assign the tumor a molecular grade. The assay was developed by comparing the gene expression profiles of grade I (i.e.,

low grade, well-differentiated) and grade III (i.e., high grade, poorly differentiated) tumors and has also been streamlined into an RT-PCR version that can also use FFPE samples. The test reclassifies grade II (i.e., intermediate grade) ER+ cancers into high or low grade categories and thereby confers significantly different prognoses on otherwise similar tumors. High GGI is associated with decreased relapse-free survival in patients who do not go on to receive adjuvant chemotherapy and is also associated with increased sensitivity to neoadjuvant chemotherapy in both ER− and ER+ patients [23].

MicroRNAs (microRNAs) in diagnosis

Lauren Chen etal suggested that microRNA may serve as potential diagnostic and prognostic biomarkers and therapeutic targets for breast cancer. Quantitative real-time PCR (qRT-PCR) array analyses of microRNAs in sera from four pairs of recurrent and non-recurrent breast cancer patients were performed. Those differentially expressed microRNAs were verified in serum samples from 42 breast cancer patients. High serum levels of miR-134 and miR-483-5p were found to be associated with some aggressive tumor behaviors. Kaplan-Meieranalysis of four up regulated microRNAs (miR-134, miR-483-5p, miR-493-3p and miR-139-3p) indicated that serum level of miR-134 can predict tumor recurrence in breast cancer patients after primary treatment. Identification of new blood biomarkers for prediction of recurrence may significant implication for breast cancer follow-up care and treatment [24].

PLAC1

Hongyan *et al.*, showed Placental-specific protein 1 (PLAC1) is an X-linked trophoblast gene that is re-expressed in several malignancies, including breast cancer, and is therefore a potential biomarker to follow disease onset and progression. Sera from 117 preoperative/pretreatment breast cancer patients and 51 control subjects, including those with fibrocystic disease, were analyzed for the presence of PLAC1 protein as well as its expression by IHC in tumor biopsies in a subset of subjects. Serum PLAC1 levels exceeded the mean plus one standard deviation (mean+SD) of the level in control subjects in 67% of subjects with ductal carcinoma in situ (DCIS), 67% with HER2+ tumors, 73% with triple-negative cancer and 73% with ER+/PR+ tumors [25].

American Society of Clinical Oncology (ASCO) Recommendations

ASCO recommendations for when to use different testing options to find out whether a person might benefit from adjuvant systemic therapy. Adjuvant therapy is a term used to describe any treatment given after breast cancer surgery. Systemic therapy is treatment that is delivered through the body's bloodstream. These tests depend on the tumor's ER/PR and HER2 results and whether the cancer has spread to lymph nodes.

People with ER/PR-positive, HER2-negative breast cancer that has not spread to the lymph nodes. Oncotype DX,EndoPredict,Breast Cancer Index (BCI),PAM50,uPA and PAI-1.

Mamma Print, for those with a high risk of cancer recurrence based on how much ER/PR is in the tumor and how quickly the cancer grows.

For people with HER-2 positive or "triple negative" (ER/PR-negative and HER2-negative) breast cancer. The tests listed above have not yet been shown to be useful for predicting risk of recurrence in people with these specific types of breast cancer. Therefore, none of these tests are currently recommended for breast cancer that is HER2 positive or triple negative [26].

Genetic Panels in Inherited Breast Cancer (Biomarkers being evaluated)
p53

p53 is the most commonly mutated gene in human cancers. Individuals who have germline mutations in TP53 have Li-Fraumeni syndrome. Patients with Li-Fraumeni syndrome are at high risk for early-onset breast cancer. The primary limitation of performing screening for germline p53 mutations is their rarity [27].

AT mutated (ATM) gene

Ataxia Telangiectasia (AT) is caused by mutations in the AT mutated (ATM) gene which leads to the generation of defective AT protein [28]. The normal AT protein detects DNA strand breaks, recruits proteins to fix the break, and prevents a cell from making new DNA until the repair is finished. People with AT are at an increased risk of multiple cancers, including lymphoma, leukemia, and breast cancer. Compared to the general population, women who are heterozygous or homozygous for AT have double the risk of developing breast cancer [29]. The relative infrequency of the mutation limits its justification for screening the general population to identify individuals at increased risk.

Phosphatase and Tensin (PTEN) gene

Mutations in the Phosphatase and Tensin (PTEN) gene can contribute to the development of a variety of cancers, including breast cancer [29] Approximately 50% of breast cancers have loss of PTEN expression, which is associated with lymph node metastases and poor survival [30]. Individuals with Cowden's disease, who have germline mutations in PTEN, have a 25–50% lifetime risk of developing breast cancer [31].

Multiple Gene Analyses

There are a variety of hereditary breast cancer

syndromes which have genetic mutations associated with them, and confer an increased risk of developing breast +/− other malignancies [32]. These include Hereditary Breast and Ovarian Cancer syndrome, with mutations in BRCA1 or BRCA2, Li-Fraumeni, with mutations in TP53, Cowden's syndrome, involving PTEN, Hereditary Diffuse Gastric Cancer syndrome, involving CDH1, Peutz-Jeghers, involving STK11, Lynch syndrome, involving MLH1, MSH2, MSH6, or PMS2, and Fanconi anemia, involving PALB2. Lifetime risk of breast cancer is over 20% for mutation carriers of these syndromes, ranging up to 80% for BRCA1 mutation carriers [32]. Gene panels have been developed to evaluate patient samples for alterations in some or all of these genes.

BROCA

This panel from the University of Washington evaluates mutations in genes involved with a variety of human cancers. BROCA is most useful for analyzing patients with a suspected cancer predisposition. An advantage of the BROCA gene panel is that specific gene testing can be selected or the investigator can opt for the entire panel. The number of genes in the panel changes over time based on new information [33].

Breast Next

This 17 gene panel developed by Ambry Genetics is very similar to the BROCA panel in that it analyzes cancer risk and is best suited for patients with a suspected hereditary predisposition to breast or ovarian cancer. Like BROCA, this panel offers the option of specific gene testing or analysis of the entire panel. A further advantage to Breast Next is that it includes duplication and deletion gene analysis [33].

BRCAPLUS

This 6 gene panel developed by Ambry Genetics performs next generation sequencing (BRCA1/2, CDH1, PALB2, PTEN, TP53). Each of the genes analyzed is linked to hereditary cancer syndromes and has published management guidelines [29].

Breast/Ovarian Cancer Panel

This 20 gene panel developed by GeneDx evaluates genes that have been linked to an hereditary disposition to breast and/or ovarian cancer [28].

Myriad myRisk® Hereditary Cancer test

Myriad myRisk® Hereditary Cancer test is a 28-gene panel that identifies an elevated risk for eight cancers (breast, ovarian, gastric, colorectal, pancreatic, melanoma, prostate, and endometrial) [28].

CONCLUSION

Molecular genomic testing will further improve in diagnosis, Prognosis and Treatment of Breast Cancer Patients.

Conflicts of Interests: There is no conflicts of intersts.

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