

The Unmet Clinical Need for New Molecular Genetic Markers in the Prognosis and Therapeutic Management of Breast Cancer

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Adenocarcinoma of the breast is a leading cause of cancer mortality among women in the United States, with an estimated 200 000 new cases and 40 000 deaths occurring each year.¹ The successful treatment of breast cancer is dependent on a number of complex factors, including the discovery of the tumor early in its course of development, the biology underlying the clinical course of the disease, and the selection and successful completion of the most appropriate treatment for an individual patient. The decision to stratify newly diagnosed breast cancer patients regarding the need for systemic adjuvant therapy has traditionally been based on the assessment of established prognostic factors, including evaluation of tumor grade, size, axillary nodes, and patient age, as well as the hormone receptor and *HER2* status.² However, it is increasingly clear that prognostic factors currently in use do not provide sufficient information to allow accurate individual risk assessment for node-negative breast cancer patients, emphasizing the need for new prognostic and therapeutic strategies for early-stage disease.^{3,4} This is particularly true in light of the rapid development of

new, targeted cancer treatments that require companion tests to predict their effectiveness⁵ and the development of new prognostic tests using genomic markers. In this regard, a greater understanding of the molecular and cellular basis of breast cancer phenotypes, and how this pertains to clinical progression and aggressiveness of disease, is an important first step in identifying and evaluating potential new molecular targets and prognostic tests for their suitability in clinical practice.

THE CLINICAL NEED FOR NEW PROGNOSTIC AND THERAPEUTIC STRATEGIES

When a diagnosis of breast cancer is made, the immediate question of importance to the patient and her doctor is "What does this diagnosis mean for her future?" Furthermore, difficult decisions about additional therapy beyond surgery are discussed to try and determine whether adjuvant therapies of any type (hormonal, radiation, and/or cytotoxic chemotherapy) will be of any additional benefit to improve her chances for survival. Approximately one third of women with lymph node metastases at the time of surgery remain free of disease recurrence for 10 years, while one third of patients who are lymph node-negative at the time of surgery develop recurrent disease. The current challenge and unmet need in breast cancer management is to be able to identify, with a high degree of accuracy, which women from each group truly do not require additional treatment, thus sparing them the side effects and potential significant toxicities of unnecessary chemotherapeutic treatment regimens.

If robust prognostic markers that

can be applied at the time of initial diagnosis to help stratify an individual patient's risk and her likelihood for recurrence were available, these tests would improve treatment decisions. Information from these tests would be invaluable clinically in helping to optimize systemic adjuvant therapies for those patients with a high risk for recurrence, while indicating local and hormonal therapies for patients with an excellent prognosis, thus avoiding unnecessary and likely toxic treatment regimens that may be of little or no benefit. In addition, for those hormone receptor-negative patients determined to be in need of systemic adjuvant treatment, any new prognostic markers able to identify those patients requiring treatment with cytotoxic agents and to define those patients with good prognosis would be highly desirable, as none exist today.

THE BIOLOGY OF BREAST CANCER AND CLINICAL MANAGEMENT

Increasingly, the growing body of knowledge regarding the biology of breast cancer is having a significant impact on clinical management and therapeutic decision making. Since the mid-1970s, evaluation of the estrogen and progesterone receptor status has been used in the clinical management of breast cancer, both as an indicator of benefit from adjuvant endocrine therapy and as a prognostic factor for disease recurrence.⁶ In addition, profiling the amplification and overexpression of the *HER2* gene has also become a key component for defining prognosis in patients who overexpress this gene and is another important determinant in therapeutic management. The *HER2* gene encodes a 185-kd transmembrane growth factor receptor with tyrosine

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kinase activity that is involved in signaling for cellular proliferation.⁷ The *HER2* gene is amplified in up to 30% of breast cancer patients and is strongly correlated with HER2 protein overexpression, early relapse, and poor overall survival.^{8,9} In addition to portending an overall poor prognosis, HER2 overexpression is predictive of response to trastuzumab (Herceptin), a humanized monoclonal antibody targeted against the protein receptor. Trastuzumab has been shown to be effective in treating breast cancer patients with HER2 overexpression, both as a single agent⁵ and in combination with more traditional chemotherapy¹⁰; however, not all HER2-positive breast cancer patients benefit from this targeted therapy approach.^{5,10}

In addition to predicting trastuzumab responsiveness,⁵ *HER2* status may modify response to other breast cancer treatment modalities, including antiestrogens and anthracycline-based chemotherapy.^{11,12} Data from a number of clinical trials has suggested that breast cancer patients whose tumors overexpress HER2 may have a better response to treatment with doxorubicin (Adriamycin).^{13,14} Both clinical and in vitro evidence supports the concept that this is not a direct effect of HER2 overexpression, but rather the result of coamplification of topoisomerase II- α (*TOP2A*), a gene with proximity to *HER2* on 17q11.2–q12.^{15,16} Topoisomerases are enzymes present in the nucleus that regulate cellular processes such as replication and transcription, and in addition are a primary molecular target for drugs of the anthracycline class of chemotherapeutic agents.¹⁶ The results of ongoing, prospective clinical trials should shed additional light on the utility of screening for *TOP2A* expression to predict sensitivity to anthracycline-based treatment regimens.

Current consensus based on many clinical trials indicates that adjuvant polychemotherapy improves survival in breast cancer, leading to its inclusion in the treatment regimens for many women at the time of diagnosis.^{17–19} Despite the favorable impact on long-term survival, not all patients treated with chemotherapy will stand to benefit, and it would be important to accurately identify specific patients for whom it would be reasonable to

avoid the administration of these potentially toxic regimens. Patients older than 70 years with node-negative cancer smaller than 1 cm in diameter that has favorable histology are unlikely to derive any additional survival benefit from chemotherapy.^{17–19} For this group of patients, the development of accurate prognostic tests would be particularly valuable, adding confidence to the decision to forgo additional treatment beyond excision and/or radiation, while potentially identifying a small subset of patients who should receive further systemic treatment.

THE PROMISE OF GENOMICS TO FURTHER REFINE PATIENT MANAGEMENT

Despite significant research efforts and the identification of many promising putative prognostic and predictive markers, the results to date have been somewhat disappointing in that there is no single tumor biologic factor available for clinical use that can accurately predict the clinical course of disease or chemotherapy response. This is likely related to the significant molecular complexities that underlie the heterogeneous biologic and clinical behavior of breast cancers. In addition to the current limitation of clinically useful biomarkers, available effective therapies are also somewhat limited for many breast cancer patients in need of systemic treatment. However, given the significant biologic heterogeneity of breast cancer, it is likely that there are subsets of patients who will derive significant benefit from cytotoxic regimens, while others will not. Accumulating evidence suggests that chemotherapy may be more targeted than previously appreciated,^{13–16} and genomics may provide more objective selection criteria to help predict which patients are likely to derive benefit from selective cytotoxic agents.

A number of authors have suggested that examination of combinations of genomic markers (biomarkers) will have significantly greater prognostic power than any single biologic factor and will bring us closer to the goal of developing clinically useful diagnostic assays, which can be used to individually tailor treatment.²⁰ The challenge is to identify the best combination of biomarkers for further investigation and valida-

tion in clinical studies. A prerequisite would be the ability to reliably and reproducibly detect these biologic factors in routine clinical samples. Technical advances from basic science and molecular biology laboratories have brought us closer to achieving this goal. Fundamental advances in molecular biology, including expansion of our knowledge of DNA sequences and the development of powerful array technologies for analyzing and profiling gene expression, have made it possible to begin to explore the specific genetic alterations that underlie human malignancy.^{21–24} These powerful technologies are well suited to target gene discovery, but given the complexity of the output, significant cost, requirement for fresh or frozen tissue, and the lack of standardization, these methodologies are impractical in the routine clinical setting in their current formats. However, significant progress has been made in adapting genomic approaches to routine, formalin-fixed, paraffin-embedded patient samples, which will in all likelihood enable the use of many complex high-throughput technologies, linked with clinical outcomes to patient samples to accelerate new developments.²⁵ Furthermore, expression array results require both tissue and clinical validation by complementary or alternative approaches to detect gene copy number changes or altered protein expression. Techniques such as fluorescence in situ hybridization or immunohistochemistry are not only useful for the validation of molecular discovery studies, they can also be further developed into new tests using biomarkers to aid in identifying prognosis. The identification of clinically relevant biologic differences between tumors, their expression profiles, and molecular classifications resulting from such studies holds the promise for the development of more powerful predictors of clinical course, as well as confining treatment to those patients who are the most likely to benefit.^{25–28}

SUMMARY

The most clinically relevant tests to help determine the prognosis for breast cancer patients, as well as to aid in the selection of maximally beneficial treatment regimens, will con-

continue to be an area of active investigation. These types of studies continue to challenge traditional diagnostic pathology with opportunities to more closely link tumor prognosis with therapeutic decision making. The future of cancer diagnosis and treatment will continue to evolve from a better understanding of the molecular basis for a tumor's biologic and clinical behavior. The pathology community has an unprecedented opportunity to play a pivotal role in these new developments and must continue to work closely with clinicians and basic scientists to help translate and interpret molecular biologic differences between tumors within a morphologic and clinical context. This understanding will most certainly lead to the development of new diagnostic assays, as well as new and novel targeted therapeutic strategies. In turn, we will potentially achieve the goal of tailoring individual treatment regimens for newly diagnosed breast cancer patients to maximize the benefit from therapy—for every patient.

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