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Professional paper

The pathologist – a key person in a multidisciplinary team of breast cancer

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Summary

Diagnosis and treatment of breast cancer has progressed rapidly in recent 20 years. The diagnosis was first based on clinical appearance that changes after the introduction of mammography in the diagnosis. Development of radiological techniques has led to the detection of a small and non-palpable lesions, and surgeons are increasingly applied conserving procedures for breast cancer. Therefore, today is a very important multidisciplinary team in the treatment of patients with breast cancer. A pathologist is a key member of the multidisciplinary team because must determine the number of prognostic and predictive factors for patients with breast cancer which requires some standardized protocols and processing of tissue samples.

INTRODUCTION

Breast cancer is the most common cancer among women in both developing and developed regions in the world. Clinical cancer develops over a long period of time, requires multiple molecular alterations, and involves evolution of cellular populations with increasingly aggressive phenotypic characteristics *(1)*. Although the time required for the process of carcinogenesis is not well established for any human cancer, estimate suggest that this multistep process unfolds over many years and possibly several decades. Breast cancer represents a diverse collection of malignant diseases of the breast with highly variable clinical behaviors and disparate response to therapy *(2)*.

Personalized oncology is evidence-based, individualized medicine that delivers the right care to the right cancer patient at the right time and results in measurable improvements in outcomes and a reduction on health care costs. The essence of personalized oncology lies in the use of biomarkers. The biomarkers can be from tissue, serum,urin or imaging and must be validated. Also, their have different importances: predictive, prognostic and early response biomarkers.

The diagnosis and treatment of breast cancer has rapidly evolved over the past 20 years. In the first part of the 20th century, treatment of breast cancer consisted of radical mastectomy, but adjuvant systemic treatment and adjuvant radiotherapy did not play a major role. Diagnosis of breast cancer was mostly made based on clinical presentation, later aided by mammography and often combined with frozen section pathology confirmation. Starting in the 1980s, there have been important alterations in the diagnosis and treatment of breast cancer, having an important impact on the diagnostic procedure employed by pathologists.

Histopathological prognostic and predictive factors

Histopathological features play an important role in guiding the treatment decisions. In addition, genetic research is starting to have an increasing impact on guiding therapy by providing prognostic and predictive factors *(3)*.

To obtain optimal morphology in the histology sections, and to obtain optimal immunohistochemical staining results, the resection specimen should be cut into thin slices immediately after surgery.

For microscopic examination the pathologist should be obtained and processed for paraffin sections full diameter of the tumour and its surroundings, small part of the tumour to perform immunohistochemistry, if there are macroscopical or radiological abnormalities in the tissue surrounding the invasive tumour, these areas should be sampled. If the surrounding tissue is without abnormalities, it is necessary to take at least two sections from macroscopically normal breast tissue.

On slides stained with hematoxylin eosin (H.E.), pathologist must determine the prognostic and predictive factors for breast cancer. This includes the histological type of cancer *(4)*, the degree of tumour differentiation and nuclear grade *(5)*, mitotic counts, peritumoral lymphovascular invasion, estrogen and progesterone receptors *(6)*, protein HER-2 *(7)* and proliferative index Ki–67 *(8)*.

Molecular markers as prognostic factors

Receptors are determined by immunohistochemistry and the results are expressed as the percentage of positive cells and intensity of staining. Staining for estrogen and progesterone receptor is allways nuclear in localization and in most institutes all patients with a tumour in which more than 10% or more 1% of the tumour cells show positive staining regardless of the intensity of staining are candidates for adjuvant hormonal therapy. According to the consensus of the St Gallen 2013. cut-off of the progesterone receptors is 20%. This value best separating luminal type A from luminal type B breast cancer. Values below 20% indicate that the progesterone receptors are negative or low *(9)*. When negative staining for estrogen and/or progesterone receptor is seen, it is important to confirm that staining of the hormone receptor-negative case has been successful. This can usually be tested, since the majority of normal breast tissues contain some nuclei ducts and lobules that are positive for estrogen and progesterone receptor. If no normal breast epithelial cells are found to show positive staining, the hormone receptor assays should be repeated on another tumour block.

HER-2 gene amplification is observed in 15–30% of invasive breast cancers and leads to HER-2 receptor overexpression. HER-2 positive invasive breast cancers respond favourable to therapies that specifically target the HER-2 protein, therefore it is very important today to identify candidates for this type of targeted therapy. Several technologies are available for determining HER-2 status, but the two most commonly used are immunohistochemistry (IHC), which measures HER-2 protein expression and CISH (chromogen in situ hybridisation) which detects HER-2 gene amplification a method that is often used today in the pathology than FISH (fluorescence in situ hybridisation). The interpretation of the results is based on the intensity and percentage of stained cells. The most commonly used score system is 0, 1+ (negative results), 2+ and 3+ (positive results). A 2+ is considered equivocal and should be followed by retesting by CISH. Women with IHC 3+ tumours are candidates for therapy with trastuzumab, but women with 2+ tumour should be retested and if the results show amplification of gene of those are candidates for trastuzumab. To ensure the highest possible accuracy, pathology centers must standardise methodologies and testing procedures.

Proliferative index is also very important and is determined by immunohistochemistry by monoclonal antibody Ki-67. Positive reaction is nuclear reaction and are counted positive nuclei in 1000 tumour cells on the high magnification and the results obtained is expressed as a percentage of positive nuclei. According to St Gallen consensus cut of value is 20% of positive cells, which means that below this value is low and value above 20% is high proliferative index *(9)*.

Based on the receptors, HER-2 status and proliferative index breast cancers are classified immunophenotypically into five subgroups: luminal type A, luminal type B HER-2 negative, luminal type B Her-2 positive, HER-2 positive (non-luminal type) and triple negative tumours. Based on the immunophenotype of the cancer patients receive appropriate therapy. The multi-gene testing remains inaccesible for the majority of women with early breast cancer, therefore is adopted clinico-pathological testing, now expressed in surrogate IHC-based classification.

In the widest sense, post-therapy effects include morpholgical and biological alterations in cancer and normal tissue after any treatment. The patients have no or little response, and the majority has a partial response to therapy. The extent of this response is associated with outcome. Identifying stage after treatment is important and provides additional prognostic information. The response in lymph nodes has more prognostic importance than does response in the breast. Small metastases after treatment, including isolated tumour cells, are representative of an incomplete pathological response *(10)*.

The role of tumor enviroment

However, the tumour microenviroment has been recognized to play an important role in oncogene-addicted tumours as well as the success of therapeutic responses to

targeted agents. In cancer therapeutics, the immune system has been shown to be important through its contribution to the efficacy of cancer therapeutics. The role of the immune system in HER-2 positive breast cancers is largely undefined, as breast cancers have long been thought not to be amenable to immune approaches. However, lymphocytic infiltration, in most cases representing a T-cell infiltrate, is correlated with better clinical outcomes in an increasing number of large breast cancer datasets. The quantity of lymphocytic infiltration assessed in tumours at baseline has also been associated with increased sensitivity to chemotherapy (higher pathological complete response rates) *(11)*. Similarly, breast cancer that are of the triple-negative subtype also have a much better outcome if they have high levels of lymphocytic infiltration at diagnosis and if they receive anthracycline-based adjuvant chemotherapy *(12)*. If patients have HER-2 positive breast cancer and infiltration of lymphocytes in a tumour, the finding of tumor infiltrating lymphocytes (TILs) can not be recommended as a guide who should or should not receive trastuzumab, but they may ultimately help define a good prognostic group of patients that may not require further addition to trastuzumab or may predict increased benefit from T-cell checkpoint inhibitors and other immunotherapies *(11)*. The simplest method of determining lymphocytic infiltration is on haematoxylin and eosin slides. It is still a semiquantitative method based on a pathologists subjective assesment *(13)*.

We investigated the expression of MAGE family gene in invasive ductal breast cancers. Their products encompass tumour associated antigenes (TAAs) recognised by human leucocyte antigen (HLA)- restricted specific T-cells.These genes, especially their products are of particular interest in tumor immunology *(14)*. By using a monoclonal antibody specific for MAGE family gene products, we have studied the expression of these TAAs in group of 144 patients with invasive ductal breast cancers. Immunohistochemical data were correlated with pathohistological prognostic factors, and the expression of MAGE gene products was typically detectable in poorly differentiated invasive ductal breast cancer. The data also support the concept of specific immunotherapy procedures targeting MAGE family TAA in highly aggressive forms of breast cancerv *(15)*.

CONCLUSION

Numerous studies in recent years have identified many prognostic and predictive factors for breast cancer. Most of them determined pathohistologically, which resulted in a large responsibility for pathologists. In addition, pathologist has become a key person in a multidisciplinary team of breast cancer and the person very responsible for the implementation of specific individual therapy.

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