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Precision Molecular Pathology of Breast Cancer

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Ashraf Khan, MD, FRCPath · Ian O. Ellis, MD, FRCPath · Andrew M. Hanby, BM, FRCPath
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Editors

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Editors
Ashraf Khan, MD, FRCPath
Department of Pathology
University of Massachusetts Medical School
UMassMemorial Medical Center
Worcester, MA
USA

Ediz F. Cosar, MD
Department of Pathology
University of Massachusetts Medical School
UMassMemorial Medical Center
Worcester, MA
USA

Ian O. Ellis, MD, FRCPath
Department of Histopathology
University of Nottingham
Nottingham
UK

Emad A. Rakha, MD, FRCPath
Department of Histopathology
University of Nottingham
Nottingham
UK

Andrew M. Hanby, BM, FRCPath
Leeds Institute of Cancer and Pathology
St. James University Hospital
Leeds, West Yorkshire
UK

Dina Kandil, MD
Department of Pathology
University of Massachusetts Medical School
UMassMemorial Medical Center
Worcester, MA
USA

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Preface

The past decade has seen an immense growth in our understanding of the molecular basis of cancer, which has made a significant impact on how we manage cancer in this era of personalized medicine. Breast cancer, which is the most common malignancy in women in the western world, has been the vanguard in the application of molecular pathology in its management. Advances in molecular pathology have led to the development of new ancillary studies that are now standard clinical practice for profiling of breast tumors permitting the tailoring of adjuvant treatment. The investigations include diagnostic and predictive biomarkers determined both by immunohistochemistry and more traditional molecular pathology techniques such as FISH. At the advancing research front further potential new targets of therapy within the molecular pathways underpinning current practice are being revealed.

With the fast pace of growth in our knowledge, practicing physicians, including pathologists, are increasingly expected to have a sound understanding of both traditional morphology based interpretation and the molecular pathology of breast cancer. Pathologists are consultants to their clinical colleagues for managing patients with breast cancer, and the role of molecular pathology has become critical in this era of personalized medicine and multidisciplinary cancer care. It is therefore important for pathologists to be familiar with advances in molecular pathology of breast cancer, so they can provide a better, informed, opinion when discussing cases with their clinical colleagues.

This book, which is part of the molecular pathology of cancer series, was put together with the aim of combining histopathologic and cytomorphologic features with changes at the molecular level, and how these latter alterations can play a role in breast cancer management. The editors are experienced practicing diagnostic breast pathologists who apply these molecular pathology techniques routinely in their practice. With the exception of one chapter where we have invited breast radiologist and medical physics experts to write on the molecular basis of breast cancer imaging, all the authors in addition to diagnostic pathologists, include cancer biologists, who focus on the molecular biology of the breast cancer. The editors, who are also the senior author on each chapter, are internationally recognized

breast pathologists who bring their own valuable insights into the interface between morphology and molecular pathology.

We are very grateful to all the contributors who have taken time out of their busy schedules to write these chapters. We would also like to take this opportunity to thank the series editor Dr. Philip Cagle for inviting us to write this book and the editorial staff at Springer Publications for all their assistance in making this project possible.

Ashraf Khan
Ian O. Ellis
Andrew M. Hanby
Ediz F. Cosar
Emad A. Rakha
Dina Kandil

Contents

Contributors

Mohammed A. Aleskandarany Department of Histopathology, Division of Cancer and Stem Cells, School of Medicine, The University of Nottingham and Nottingham University Hospitals NHS Trust, Nottingham City Hospital, Nottingham, UK

Jennifer L. Clark Department of Pathology, University of Massachusetts Medical School, UMassMemorial Medical Center, Three Bioetch, One Innovation Drive, Worcester, MA, USA

Ediz F. Cosar Department of Pathology, University of Massachusetts Medical School, UMassMemorial Medical Center, Worcester, MA, USA

Ian O. Ellis Division of Cancer and Stem Cells, Department of Histopathology, School of Medicine, University of Nottingham and Nottingham University Hospitals NHS Trust, Nottingham City Hospital, Nottingham, UK

Yuna Gong Department of Pathology, University of Massachusetts Medical School, UMassMemorial Medical Center, Three Biotech, One Innovation Drive, Worcester, MA, USA

Andrew M. Hanby St. James University Hospital/University of Leeds, Leeds Institute of Cancer and Pathology (LICAP), Leeds, West Yorkshire, UK

Thomas A. Hughes Leeds Institute of Biologial and Clinical Sciences (LIBACS), St. James University Hospital/University of Leeds, Beckett Street, Leeds, West Yorkshire, UK

Lloyd Hutchinson Department of Pathology, University of Massachusetts Medical School, UMassMemorial Medical Center, Worcester, MA, USA

Dina Kandil Department of Pathology, University of Massachusetts Medical School, UMassMemorial Medical Center, Three Biotech, One Innovation Drive, Worcester, MA, USA

Andrew Karellas Department of Radiology, University of Massachusetts Medical School, Worcester, MA, USA

Ashraf Khan Department of Pathology, University of Massachusetts Medical School, UMassMemorial Medical Center, Three Biotech, One Innovation Drive, Worcester, MA, USA

Arthur M. Mercurio Department of Molecular, Cell and Cancer Biology, University of Massachusetts Medical School, Worcester, MA, USA

Rebecca A. Millican-Slater Department of Histopathology, Leeds University Hospitals NHS Trust, Leeds, UK

Marjan Mirzabeigi Department of Pathology, University of Massachusetts Medical School, UMassMemorial Medical Center, Worcester, MA, USA

Abir A. Muftah Department of Histopathology, Division of Cancer and Stem Cells, School of Medicine, The University of Nottingham and Nottingham University Hospitals NHS Trust, Nottingham City Hospital, Nottingham, UK

Abhik Mukherjee Division of Cancer and Stem Cells, Department of Histopathology, School of Medicine, University of Nottingham and Nottingham University Hospitals NHS Trust, Nottingham City Hospital, Nottingham, UK

Claire Nash St. James University Hospital/University of Leeds, Leeds Institute of Cancer and Pathology (LICAP), Leeds, West Yorkshire, UK

Emad A. Rakha Department of Histopathology, Division of Cancer and Stem Cells, School of Medicine, Nottingham University Hospitals NHS Trust, Nottingham City Hospital, The University of Nottingham, Nottingham, UK

Ali Sakhdari Department of Pathology, University of Massachusetts Medical School, UMassMemorial Medical Center, Worcester, MA, USA

Sanjoy Samanta Department of Molecular, Cell and Cancer Biology, University of Massachusetts Medical School, Worcester, MA, USA

Valerie Speirs St. James University Hospital/University of Leeds, Leeds Institute of Cancer and Pathology (LICAP), Leeds, West Yorkshire, UK

James L. Thorne Department of Mathematical and Physical Sciences (MAPS), School of Food Science and Nutrition, Woodhouse Lane, University of Leeds, Leeds, West Yorkshire, UK

Srinivasan Vedantham Department of Radiology, University of Massachusetts Medical School, Worcester, MA, USA

Gopal R. Vijayaraghavan Department of Radiology, University of Massachusetts Medical School, Worcester, MA, USA

Vighnesh Walavalkar Department of Pathology, University of Massachusetts Medical School, UMassMemorial Medical Center, Worcester, MA, USA

Michelle Yang Department of Pathology, University of Massachusetts Medical School, UMassMemorial Medical Center, Worcester, MA, USA

Chapter 1
Molecular Basis of Breast Cancer Imaging

Gopal R. Vijayaraghavan, Srinivasan Vedantham, Ashraf Khan and Andrew Karellas

Introduction

Over the past decade, annually for women 50 years of age or older, the breast cancer incidence rate in the United States has ranged from 400 to 500 per 100 000 women and the breast cancer mortality rate has ranged from 60 to 80 per 100 000 women [1]. Though there has been a decline in the breast cancer mortality in the past decade it continues to be the second leading cause of death after lung cancer in women over 40 years of age.

Breast cancer continues to be a major health issue among women in the United States. Screening mammogram has significantly contributed to the reduction in mortality. However, screening mammogram has its own limitations. Its sensitivity is 80 % in fatty breasts but is substantially lower in dense breasts [2]. On average nearly 30 % of women reporting for mammograms have dense breasts and 1 in 2 cancers in dense breasts are missed on mammograms due to the masking effect caused by overlapping tissues.

Notwithstanding the limitations of screening mammograms, it is widely considered the most effective tool for the early detection of breast cancer [3], and supplementing mammography with ultrasound and MRI greatly improves the diagnosis of breast cancer. Further improvements in sensitivity and specificity for diagnosis of breast cancer are likely to involve alternative imaging approaches that address

A. Khan (✉)
Department of Pathology, University of Massachusetts Medical School, UMassMemorial Medical Center, Three Biotech, One Innovation Drive, Worcester, MA 01605, USA
e-mail: Ashraf.Khan@umassmemorial.org

G.R. Vijayaraghavan · S. Vedantham · A. Karellas
Department of Radiology, University of Massachusetts Medical School, Worcester, MA 01605, USA

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the limitations of existing imaging modalities or provide for imaging new contrast mechanisms. An example of the former is digital breast tomosynthesis [4], which can reduce the tissue overlap observed with mammography.

Mammograms and ultrasound images represent anatomic abnormalities that are associated with cancer. Magnetic Resonance Imaging (MRI) with injected contrast media better depicts the physiology of the tumor due to enhancement of tumor-associated angiogenesis and this partly explains its higher sensitivity compared to mammography. Changes at the cellular level that distinguish cancerous cells from benign breast tissue have been explored in newer, innovative imaging techniques. Some of the imaging techniques described are still experimental and not yet considered 'standard of care'. Some of the changes at the cellular level include new blood flow, angiogenesis, expression of protein receptors in breast cancer cells resulting in increased uptake of specific ligands and changes in oxy or deoxy-hemoglobin content of the tumor cells. Radionuclides and optical probes that target specific proteins in the cells are being investigated. Some of these newer modalities can be combined with traditional imaging as part of multimodal imaging to further improve our diagnostic capability [5].

Over the past decade advances in imaging and instrumentation have helped establish molecular breast imaging (MBI) as a useful supplemental tool [6–8]. Cost constraints, tumor size resolution, radiation dose, and sparse availability were some of the cited reasons why these modalities have not gained widespread acceptance [9]. Many of these issues continue to be addressed. Radiation dose from radionuclide-based molecular imaging [10] continues to remain a major impediment compared to established screening mammogram.

Molecular imaging reflects both tumor morphology and physiology and thus has some inherent advantages over conventional mammograms, particularly in a situation of radiographic dense breasts.

MBI techniques currently available include:

1. Breast-specific gamma imaging (BSGI).
2. Positron emission mammography (PEM).
3. Optical imaging with near-infrared spectroscopy.

In addition, imaging tests that can be performed in ex vivo specimens to evaluate cancer margins include:

1. Optical imaging with confocal microscopy.
2. Terahertz imaging.

Breast-Specific Gamma Imaging (BSGI)

While mammography and ultrasound rely on anatomical changes in the breast (calcifications, masses, architectural distortion, or asymmetry), BSGI relies on the physiology (blood flow, mitochondrial activity, and angiogenesis) of the tumor to make the diagnosis.

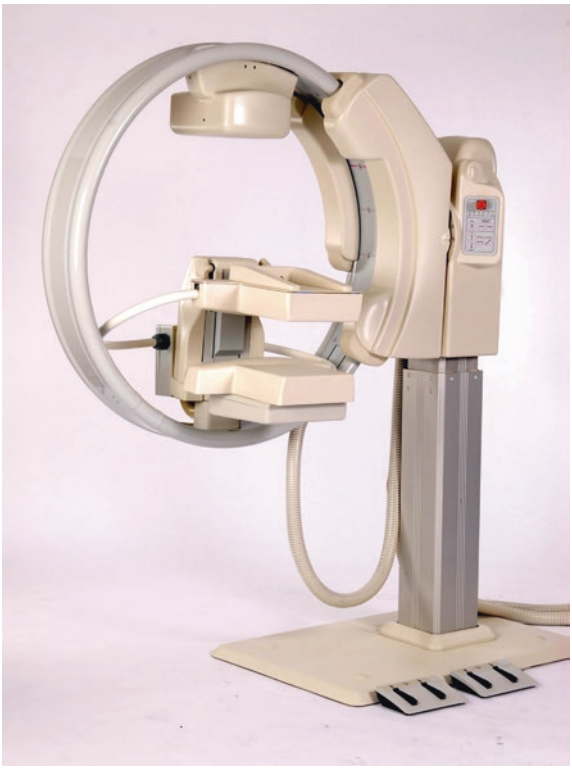

Fig. 1.1 Picture of a molecular breast imaging system. *Courtesy of Jason Koshnitsky, Gamma Medica, Inc., Salem, NH*

A standard two-view mammogram continues to be the gold standard in the evaluation of breast cancers, notwithstanding recent controversies [11]. The sensitivity of mammogram is limited [2]. In dense breasts, small cancers are hidden and can be missed [12, 13]. Breast ultrasound and tomosynthesis have shown the ability to detect some non-palpable breast cancers beyond mammography and address this limitation to a large extent [14, 15]. BSGI also known as molecular imaging of the breast has higher negative predictive value for breast cancers. Figure 1.1 shows a picture of MBI system.

Indications

The indications for performance of a BSGI study include high-risk surveillance, alternative to MRI, palpable breast masses with a negative mammogram and ultrasound, a newly diagnosed breast cancer with occult foci, and in women with breast implants or following direct silicone injection to resolve a difficult question.

Initially studies were performed on a conventional gamma camera and hence there were issues related to optimal positioning of the breast and poor resolution; this technique did not detect small cancers less than 1 cm. The sensitivity was

less than 50 %, making it a less attractive alternative [6, 9]. Over the last 20 years advancements in gamma ray detector technology (for example, the use of the semiconductor cadmium zinc telluride) and the use of dedicated dual head breast scanners have improved both energy and spatial resolution. This has enabled detection of tumors as small as 1–3 mm. Also, production of images that are oriented similar to standard mammograms has made it easier for radiologists to interpret these studies. These improvements have also resulted in decreased radiopharmaceuticals doses, making the test more acceptable [7, 16].

Technique

MBI uses the radiotracer technetium (^{99m}Tc) sestamibi in doses of about 20 mCi (740 mBq) injected intravenously (IV) in one of the antecubital veins. Imaging starts immediately and continues up to the desired number of counts per image, approximately 100,000. Images are obtained with breast oriented in a manner similar to the standard mammogram; craniocaudal (CC) and mediolateral (MLO) images of both breasts. The image acquisition time is about 10 min for each view, to a total acquisition time of 40 min for a complete study [9, 17]. The breast compression is less than that in a mammogram (a pressure of 15 lbs/square in. as opposed to 45 lbs/square in. on a conventional mammogram).

Advantages

In addition to its utility as an adjunct diagnostic tool, MBI is an attractive imaging test from the cost point of view because of the wide availability of the radiopharmaceutical and compact size of the imaging equipment. It is a useful problem-solving tool particularly in patients unsuited for MRI, because of metallic implants, renal dysfunction, or claustrophobia. BSGI has high sensitivity, specificity, and positive predictive value (PPV) compared to standard mammograms and ultrasound evaluation [7–9, 16–18]. Figure 1.2 shows an example. Weigert et al. [8] in a multicenter study determining the impact of molecular imaging concluded that statistically BSGI was more accurate (better sensitivity, specificity and PPV) than ultrasound, when findings were discordant from a standard mammogram. Lately, BSGI guided biopsy systems [6, 8] have become available which allows confirmation of pathology results and better correlation with imaging findings.

Limitations

Poor visualization of chest wall and axilla are some of the drawbacks, which can be overcome with additional views. The inability to obtain all of the breast tissue in the field of view (FOV) to a large extent has been alleviated by offering nuclear medicine technologists training in breast positioning by mammographers [9]. In view of potential radiation risks the dose of the injected radiopharmaceutical has

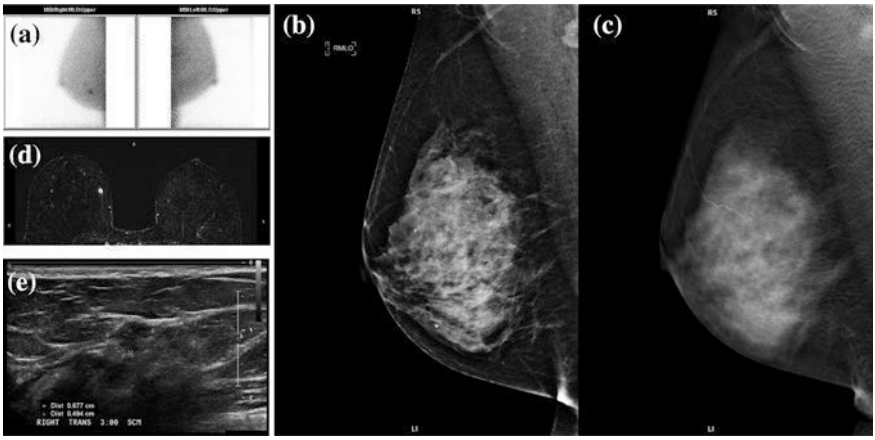

Fig. 1.2 An asymptomatic postmenopausal woman with a prior negative mammogram participated in a research study evaluating the effect of caffeine on Tc99 m sestamibi uptake. Her molecular breast imaging (*MBI*) exam (bilateral MLO) was positive (**a**). There was a focal area of moderate intensity radiotracer uptake in the lower inner right breast at middle depth measuring 1 × 0.7 × 0.8 cm. Subsequent digital mammogram (**b**), and digital breast tomosynthesis (**c**) showed no correlate to the MBI finding. Breast MRI (**d**) depicted a 0.6 × 0.6 × 0.9 cm enhancing round mass with slight irregular margins corresponding to the abnormality identified by MBI (**a**). Second look ultrasound (**e**) followed by ultrasound guided biopsy indicated a 6 mm invasive ductal carcinoma. *Courtesy of Michael K. O'Connor, Ph.D., Mayo Clinic, Rochester, MN*

been steadily decreasing. Initial trials on BSGI used doses of 30 mCi (1110 mBq), currently this has dropped to 20 mCi and some centers use only 10–15 mCi of ^{99m}Tc [7]. Trials at Mayo Clinic in Rochester, MN are experimenting with a dose as low as 4 mCi and by enhancing image quality with digital post-processing. At this dose the radiation dose to the breast is comparable to a standard two-view mammogram. The results from this study will determine if MBI has a role in screening, particularly for the intermediate risk category, where the benefit of MRI is not clearly defined. While one of the earlier limitations of BSGI was the poor sensitivity of molecular imaging in identifying sub-cm tumors, with recent advances, Hruska et al. [16] demonstrated sensitivities up to 80 % for tumors less than 1 cm.

Positron Emission Mammography (PEM)

Positron emission tomography (PET) imaging is a useful diagnostic test in many malignancies, but has not been accepted as standard of care in breast cancers [19]. Dedicated PEM scanners for breast have been developed, providing higher resolution than whole body PET scanners [20]. In PEM and PET imaging, the radiotracer fluoro-deoxyglucose (18-FDG) is used, providing a physiological measure of increased metabolic activity. While a promising tool it also demonstrates 'hot spots' at inflammatory and infective sites resulting in false positives.

Limitations

In order for the test to be sensitive, it is essential that there is good regulation of glucose levels and the blood glucose levels must be below 120 mg/dl. In order to achieve sufficient gamma counts in the image it is necessary for the patient to wait at least 2 h after administration of the radiopharmaceutical before imaging the breast. Also, it is important for the patient to remain quiet and warm to prevent 'hot spots' from unusual muscular activity. One of the limitations of the earlier whole body PET scanners was its inability to pick up sub-cm cancers. This has been addressed to a large extent by the development of new, dedicated breast scanners. Also, low grade tumors and some invasive lobular cancers and ductal carcinoma in situ do not show avid uptake of the radiotracer [19]. Radiation concern continues to be a major limitation [10]. PEM suffers from some of the same limitations associated with positioning as noted in BSGI. Not only does the imaging take 40 min (10 min for each view), but the patient needs to wait about 2 h post tracer injection before the images can be obtained. Like BSGI, the sensitivity in PEM imaging showed a declining trend with smaller sized tumors. In addition, PEM equipment is more expensive than BSGI and requires access to the 18-FDG radiotracer. Hence, PET and PEM are available only in a limited number of clinical practices in the United States.

Advantages

The availability of more recent prototypes of dedicated PEM scanners with its ability to perform imaging guided biopsies has made it an attractive additional imaging tool [19, 21]. PEM sensitivity matched MRI for single lesions and the sensitivity for unsuspected lesion was around 85 % [22, 23]. PEM had higher specificity for unsuspected lesion compared to MRI. PEM imaging is useful in identifying the extent of the tumor and staging, evaluating response to treatment, identifying sites of distant metastases, and distinguishing a scar from recurrence [19, 20, 22, 23].

Research studies over the past 10–15 years have established the role and value of MBI and PEM in breast imaging. While the newer breast molecular imaging modalities have shown promise, they are still only useful as supplemental imaging tools that can increase the radiologists' confidence in detecting breast cancer and cannot supplant established modalities such as screening mammogram, ultrasound, and MR imaging. Additional regulatory approvals are needed for the clinical site to handle radioactivity.

Radiation Risks

Since molecular imaging involves substantial radiation dose to part of the body other than the breast, there is concern about risk of cancer for radiosensitive organs; in the urinary bladder with PEM studies and in the colon with BSGI

studies. Hendrick [10] has estimated the lifetime attributable risk (LAR) of a fatal cancer in BSGI studies at standard recommended doses of 20–30 mCi (740–1100 MBq) to be 20–30 times that of a digital mammogram in woman aged 40 years, and 23 times higher with a single PEM study at standard 10 mCi (370 MBq) dose of 18-FDG.

It is also relevant to add that even though considerable advancements have been made in radiotracer-based molecular imaging of the breast, currently it is not a screening tool. Its primary role may be as an adjunct to standard mammography and ultrasound, particularly in women with dense breasts and in the intermediate risk category. In the high-risk women, MRI with its proven track record as the modality with the highest sensitivity for detection is the established modality. Both BSGI and PEM/PET have the advantage of identifying physiological changes that distinguish a cancerous lesion from benign tissue and also identify additional foci in the ipsilateral and contralateral breast. They are also helpful imaging options to monitor response to chemotherapy drugs. Their sensitivity is however known to decrease with smaller sized tumors. A higher incidence of false positive tracer uptake has been noted in fibrocystic lesions, growing fibroadenomas, and fat necrosis. PEM has shown to be useful in distinguishing a scar from recurrence where conventional imaging findings are equivocal.

Optical Imaging with Near Infrared Spectroscopy

Transillumination of the breast using light, referred to as diaphanography, was proposed in 1920s. Variants of this approach were investigated till the early 1990s. However, the approach was not recommended for breast cancer screening [24]. Better understanding of the contrast mechanisms, characterization of absorption and scattering properties of breast tissues at various wavelengths, and techniques for modeling optical photon transport through tissues have facilitated development of quantitative methods for diffuse optical spectroscopic imaging. Diffuse optical imaging using continuous wave, time domain, or frequency domain measurements at near-infrared (NIR) wavelengths can be used to provide noninvasive in vivo quantitation of attenuation and scattering properties of breast tissue. This can be used to determine total hemoglobin content, oxygen saturation (ratio of oxygenated hemoglobin to total hemoglobin), water, and lipid content. Extension of the approach to 3D imaging, similar to computed tomography (CT), has resulted in diffuse optical tomography (DOT) systems. Hand-held diffuse optical spectroscopy imaging systems have been developed and continue to be refined [25, 26]. Stand-alone DOT prototype systems for adjunctive use in diagnostic breast imaging have been developed by academic investigators [27, 28] and by commercial entities (SoftScan®, Advanced Research Technologies, Inc., Montreal, Canada; CTLM®, Imaging Diagnostic Systems, Inc., Fort Lauderdale, USA).

In a study of 90 subjects, DOT showed that the ratio of total hemoglobin in the abnormality to that in the normal contralateral breast was statistically different for

malignant tumors [29]. However, the study noted that there may be a resolution threshold of approximately 6 mm. Development of multimodality systems combining NIRS with X-ray imaging has been reported [30–32]. In a study of 189 breasts from 125 subjects including 51 breasts with lesions, a statistically significant increase was observed for total hemoglobin in malignant tumors larger than 6 mm compared to fibroglandular tissue [33]. A hand-held probe combining ultrasound with NIR imaging has been developed, and in a study of 65 subjects with 81 lesions significantly higher concentration of total hemoglobin was observed in malignancies than benign lesions [34].

Development of NIR systems integrated with MRI [35, 36] that incorporates image information from MRI during NIRS reconstruction as well as clinical evaluation of such multimodality systems have shown that malignant tumor exhibit higher concentration of total hemoglobin and lower oxygen saturation. The use of exogenous contrast agents such as indocyanine green as well as tumor-targeted contrast agents that are under development can preferentially enhance lesions and could improve differential diagnosis. Monitoring of neoadjuvant chemotherapy response with a hand-held diffuse optical spectroscopic imaging system in a limited dataset showed that significant changes in oxygenated hemoglobin could be observed approximately 90 days after initiation of therapy [37]. In order to reduce re-excision rates following breast conserving surgery (BCS), NIR-based optical imaging systems to assess tumor margins are being investigated [38, 39]. In summary, the past two decades have witnessed substantial improvement in optical imaging techniques using NIR spectroscopy, and its transition to routine use for some clinical applications is highly probable in the near future.

Optical Imaging with Confocal Microscopy

In the management of early breast cancer, BCS is the standard surgical procedure, where excision of the least volume of breast tissue free of tumor cells at the margins is the goal. However, the current surgical literature [40] estimates positive margins at surgery in the range of 20–70 %. This necessitates revision excision. Currently, the quickest means to evaluate tumor margins is the ‘traditional frozen section’. This process is however laborious, time-consuming, does not include the entire tumor surface, and is limited by freezing artifacts of the surgical margins. Cauterization surgery also limits evaluation. Currently, there are many experimental, real-time, imaging options that are being evaluated. There is a need for such techniques to be cost-effective, reproducible, and dependable.

We performed an experimental trial [41–43] of excised lumpectomy specimens at the University of Massachusetts Medical School in collaboration with the Medical Physics department at University of Massachusetts, Lowell, MA. The investigators evaluated lumpectomy specimens from breast cancer patients with polarization techniques after staining the specimen with dilute methylene blue. Wide-field polarization for quick macroscopic survey and small FOV confocal

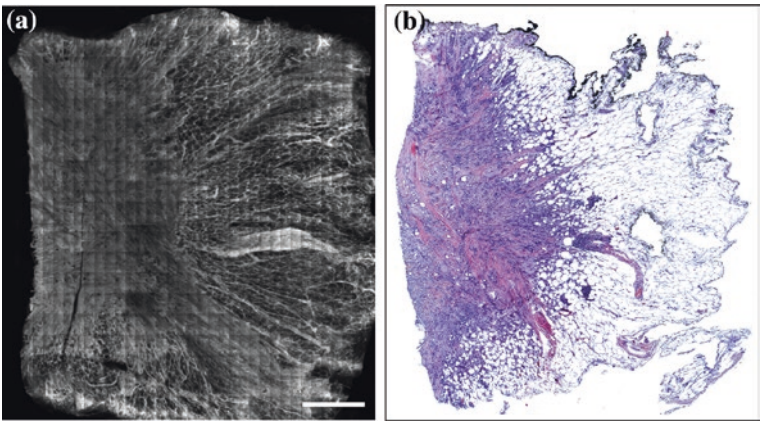

Fig. 1.3 Wide-field fluorescence polarization image (**a**) of a tissue section from a breast lumpectomy specimen with corresponding histopathology at scanning magnification (**b**) showing good demarcation between the benign (*right half*) and malignant (*left half*) breast tissue. *Fluorescence polarization image courtesy Anna Yaroslavsky, Ph.D., University of Massachusetts, Lowell, MA*

microscopy for small FOV with high resolution was performed, images analyzed, and later correlated with findings at Hematoxylin and Eosin (H&E) stained pathology slides evaluated by a trained breast pathologist (Fig. 1.3). The difference in the reflectance and fluorescence polarization values for benign and cancerous tissue was exploited. In these studies, Patel et al. [41–43] observed good correlation between fluorescence polarization values and findings on H&E stained sections of benign and malignant breast tissue on histopathology. The reflectance polarization values did not correlate as well. While the researchers concede to slight misregistration between confocal microscopy images and H&E stained specimens, the ease of use has good future potential to evaluate ex vivo specimens. The instrument could also be used in vivo on patients on the operating table to discern any residual malignant tissue that merits excision. A clinical trial is underway to evaluate the utility of this imaging technique for intraoperative evaluation of margins during BCS.

Terahertz Imaging

Accurate assessment of surgical margins of the excised breast specimen is very important in BCS in order to minimize the likelihood of re-excision. This is particularly relevant in the surgical treatment of invasive breast cancer followed by whole breast radiation therapy [44]. The reference standard for the determination of the tumor margins is by sectioning and imaging the excised specimen by conventional pathology procedures. However, more expedient techniques have been investigated over the years that allow prompt margin assessment at the intraoperative stage, thus affording the opportunity for the surgeon to excise additional tissue

if needed. Breast specimen radiography has been used for many years for this purpose. This approach is used routinely in clinical practice but it has certain inherent limitations. Radiography generates planar images of a thick three-dimensional specimen; it provides good contrast for identification of surgical margins on the basis of changes in tissue composition and density, but it is not known to differentiate well between normal tissue and cancer especially when the malignancy does not exhibit prominent morphologic changes in tissue composition and density.

Advanced three-dimensional imaging technologies such as micro-Computed Tomography and micro-MRI have been developed but are limited to research applications due to their complexity and cost. For intraoperative imaging of breast specimens, the trend in recent years has been to identify imaging approaches that can provide improved discrimination between tumor, and surrounding tissue compared to X-ray imaging. Therefore, imaging techniques that are sensitive to the molecular differences between normal and abnormal tissues may improve identification of tumor margins compared to planar X-ray imaging. Interrogation of specimens with certain types of electromagnetic (EM) radiation generates reflected or transmitted signals with intensity and spectral characteristics that may vary substantially between tumors and normal tissue. These radiations include infrared, radiofrequency, or terahertz (10^{12} Hz) radiation and their application may range from detection of the presence of abnormal tissue to assessing their invasive potential [45–47]. In the case of breast surgery, the excised breast specimen is irradiated and the returning signal after absorption, diffuse reflectance, or fluorescence in the tissue is detected and analyzed. Some techniques rely on the detection and analysis in a non-imaging approach while others generate images of the surface of the specimen, which contain intensity and spectroscopic information. One of the newest approaches uses radiation in the terahertz region of the electromagnetic spectrum; this is the part of the spectrum between infrared to microwave with a corresponding wavelength in the region of about 0.05 mm to 1 mm. This type of radiation, also called T-rays, is relatively new to biomedical applications because the development of efficient and compact sources and detectors for biomedical applications has been gradual in the past 20 years. Unlike X-rays that can easily transmit through the entire body, terahertz radiation is readily absorbed by water in the tissue and therefore transmission measurements in thick specimens are not feasible. It penetrates only a few micrometers in the breast specimen depending on the frequency used. The reflected and scattered component of terahertz radiation carries information on composition that can be used to characterize the morphology and composition of tissues. This signal can be used to form an image with compositional topography that represents its molecular status of the specimen at its surface to a depth of a few micrometers below the surface. Tissue contrast can be observed because of differences in attenuation and refractive index of the specimen and these properties have been used to assess the margins of excised breast specimens [48–52]. Therefore, imaging and spectroscopy with terahertz waves is performed in the reflection mode in a scanning beam approach. Images of the specimen can be generated at a spatial resolution, which may vary between about 0.1 and 1.0 mm depending on the imaging system and wavelength. Terahertz images

may be combined with images at other wavelengths for improved contrast and delineation of the lesion. Discrimination between normal and malignant tissues can be challenging from the raw images without proper image analysis. Some tissues, glandular and adipose for example, can be easily differentiated in terahertz imaging because of their pronounced differences in their refractive indices. It may be argued that conventional light in the visible spectrum easily discriminates between adipose and other tissues. However, terahertz appears to exhibit certain properties, which may enable detection of features that are characteristic to biochemical changes at the surface of the specimen that are associated with tumors [49].

Terahertz beam reflection, scattering, and spectra from biological specimens generally reveal variations in water composition. Under controlled conditions, Terahertz radiation can also provide characterization based on the concentration of amino acids, and proteins, and other biochemical components [53]. In the case of breast specimens, large differences in the refractive index between fibrous tissue and adipose tissue have been observed due to the large differences in their respective refractive indices and substantial differences have also been observed between fibrous tissue and breast tumors [54]. Such differences and other interactions with tissues can be used to generate images that reveal tumors in a background of healthy tissue in the specimen. In principle, other characteristics such as high levels of protein or amino acids may generate a signal under optimal conditions but at this time a complete accounting of all the components that give rise to contrast between tumors and healthy tissue has not been established.

Other radiations may be used to assess breast surgical specimen margins but at this time, specimen radiography with visual inspection is the most commonly used technique. Interrogation of the specimen with terahertz and other radiations has the potential to provide more specific information on tumor margins assuming that their refractive index and reflection properties are capable of discriminating between normal and malignant tissue. Other techniques for this purpose using optical coherence tomography with infrared radiation are being explored [55].

References

1. DeSantis C, Ma J, Bryan L, Jemal A. Breast cancer statistics, 2013. CA Cancer J Clinic. 2014;64(1):52–62.
2. Mandelson MT, Oestreicher N, Porter PL, White D, Finder CA, Taplin SH, White E. Breast density as a predictor of mammographic detection: comparison of interval and screen detected cancers. J Natl Cancer Inst. 2000;92(13):1081–7.
3. Drukteinis JS, Mooney BP, Floers CI, Gatenby RA. Beyond mammography: new frontiers in breast cancer screening. American J Med. 2013;126(6):472–9.
4. Niklason LT, Christian BT, Niklason LE, Kopans DB, Castleberry DE, OpsahlOng BH, Landberg CE, Slanetz PJ, Giardino AA, Moore R, Albagli D, DeJule MC, Fitzgerald PF, Fobare DF, Giambattista BW, Kwasnick RF, Liu JQ, Lubowski SJ, Possin GE, Richotte JF, Wei CY, Wirth RF. Digital tomosynthesis in breast imaging. Radiology. 1997;205(2):399–406.
5. Herranz M, Ruibal A. Optical imaging in breast cancer diagnosis: the next evolution. J Oncol. 2012;2012:863747.

6. Hruska CB, O'Connor MK. Nuclear imaging of the breast: translating achievements in instrumentation into clinical use. Med Phys. 2013;40(5):050901–23.
7. Rhodes DJ, Hruska CB, Phillips SW, Whaley DH, O'Connor MK. Dedicated dual-head gamma imaging for breast cancer screening in women with mammographically dense breasts. Radiology. 2011;258(1):106–18.
8. Weigert JM, Bertrand ML, Lanzkowsky L, Stern LH, Kieper DA. Results of a multicenter patient registry to determine the clinic impact of breast specific gamma imaging, a molecular breast imaging technique. AJR. 2012;198:W69–75.
9. O'Connor MK, Philips SW, Hruska CB, Rhodes DJ, Collins DA. Molecular breast imaging: advantages and limitations of a scintimammographic technique in patients with small breast tumors. Breast J. 2007;13(1):3–11.
10. Hendrick RE. Radiation doses and cancer risks from breast imaging studies. Radiology. 2010;257(1):246–53.
11. Miller AB, Wall C, Baines CJ, Sun P, To T, Narod SA. Twenty five year follow up for breast cancer incidence and mortality of the Canadian national breast screening study: randomized screening trail. BMJ. 2014;348:g366.
12. Checka CM, Chun JE, Schnabel FR, Lee J, Toth H. The relationship of mammographic density and age: implications for breast cancer screening. AJR. 2012;198:W292–5.
13. Buist DS, Porter PL, Lehman C, Taplin SH, White E. Factors contributing to mammography failure in women aged 40–49 years. J Natl Cancer Inst. 2004;96(19):1432–40.
14. Ciatto S, Houssami N, Bernardi D, Caumo F, Pellegrini M, Brunelli S, Tuttobene P, Bricolo P, Fanto C, Valentini M, Montemezzi S, Macaskill P. Integration of 3D digital mammography with tomosynthesis for population breast-cancer screening (STORM): a prospective comparison study. Lancet Oncol. 2013;14(7):583–9.
15. Giuliano V, Giuliano C. Improved breast cancer detection in asymptomatic women using 3D-automated breast ultrasound in mammographically dense breasts. Clin Imaging. 2013;37(3):480–6.
16. Hruska CB, Phillips SW, Whaley DH, Rhodes DJ, O'Connor MK. Molecular breast imaging: use of a dual-head dedicated gamma camera to detect small breast tumors. AJR. 2008;191:1805–15.
17. Brem RF, Rapelyea JA, Zisman G, Mohtashemi K, Raub J, Teal CB, Majewski S, Welch BL. Occult breast cancer: scintimammography with high-resolution breast-specific gamma camera in women at high risk for breast cancer. Radiology. 2005;237:274–80.
18. Brem RF, Floerke AC, Rapelyea JA, Teal C, Kelly T, Mathur V. Breast specific gamma imaging as an adjunct imaging modality for the diagnosis of breast cancer. Radiology. 2008;247:651–7.
19. Kalles V, Zografos GC, Provatopoulou X, Koulocheri D, Gounaris A. The current status of positron emission mammography in breast cancer diagnosis. Breast Cancer. 2013;20:123–30.
20. Narayanan D, Madsen KS, Kalinyak JE, Berg WA. Interpretation of positron emission mammograpghy and MRI by experienced breast imaging radiologists: performance and observer reproducibility. AJR. 2011;196:971–81.
21. Kalinyak JE, Schilling K, Berg WA, Narayanan D, Mayberry JP, Rai R, Dupree EB, Shusterman DK, Gittleman MA, Luo W, Matthews CG. PET guided breast biopsy. Breast J. 2011;17(2):143–51.
22. Berg WA, Madsen KS, Schilling K, Tartar M, Pisano EA, Larsen LH, Narayanan D, Ozonoff A, Miller JP, Kalinyak JE. Breast cancer: comparative effectiveness of positron emission mammography and MR imaging in presurgical planning for the ipsilateral breast. Radiology. 2011;258:59–72.
23. Berg WA, Madsen KS, Schilling K, Tartar M, Pisano ED, Larsen LH, Narayanan D, Kalinyak JE. Comparative effectiveness of positron emission mammography and MRI in the contralateral breast of women with newly diagnosed breast cancer. AJR. 2012;198:219–32.
24. Centers for Disease. C. Inappropriate use of transillumination for breast cancer screening–Wisconsin, 1990. MMWR. Morb Mortal Wkly Rep. 1991;40(18):293–6.

25. Shah N, Cerussi A, Eker C, Espinoza J, Butler J, Fishkin J, Hornung R, Tromberg BJ. Noninvasive functional optical spectroscopy of human breast tissue. Proc Nat Acad Sci. 2001;98(8):4420–5.
26. Gonzalez J, Decerce J, Erickson SJ, Martinez SL, Nunez A, Roman M, Traub B, Flores CA, Roberts SM, Hernandez E, Aguirre W, Kiszonas R, Godavarty A. Hand-held optical imager (Gen-2): improved instrumentation and target detectability. J Biomed Opt. 2012;17(8):081402–1.
27. McBride TO, Pogue BW, Jiang S, Osterberg UL, Paulsen KD. A parallel-detection frequency-domain near-infrared tomography system for hemoglobin imaging of the breast in vivo. Rev Sci Instrum. 2001;72(3):1817–24.
28. Culver JP, Choe R, Holboke MJ, Zubkov L, Durduran T, Slemp A, Ntziachristos V, Chance B, Yodh AG. Three-dimensional diffuse optical tomography in the parallel plane transmission geometry: evaluation of a hybrid frequency domain/continuous wave clinical system for breast imaging. Med Phys. 2003;30(2):235–47.
29. Poplack SP, Tosteson TD, Wells WA, Pogue BW, Meaney PM, Hartov A, Kogel CA, Soho SK, Gibson JJ, Paulsen KD. Electromagnetic breast imaging: results of a pilot study in women with abnormal mammograms. Radiology. 2007;243(2):350–9.
30. Boverman G, Fang Q, Carp SA, Miller EL, Brooks DH, Selb J, Moore RH, Kopans DB, Boas DA. Spatio-temporal imaging of the hemoglobin in the compressed breast with diffuse optical tomography. Phys Med Biol. 2007;52(12):3619–41.
31. Krishnaswamy V, Michaelsen KE, Pogue BW, Poplack SP, Shaw I, Defrietas K, Brooks K, Paulsen KD. A digital x-ray tomosynthesis coupled near infrared spectral tomography system for dual-modality breast imaging. Opt Express. 2012;20(17):19125–36.
32. Vedantham S, Shi L, Karellas A, Michaelsen KE, Krishnaswamy V, Pogue BW, Paulsen KD. Semi-automated segmentation and classification of digital breast tomosynthesis reconstructed images. In: Conference Proceeding of IEEE Engineering in Medicine and Biology Society, EMBC, 2011. p. 6188–6191.
33. Fang Q, Selb J, Carp SA, Boverman G, Miller EL, Brooks DH, Moore RH, Kopans DB, Boas DA. Combined optical and X-ray tomosynthesis breast imaging. Radiology. 2011;258(1):89–97.
34. Zhu Q, Cronin EB, Currier AA, Vine HS, Huang M, Chen N, Xu C. Benign versus malignant breast masses: optical differentiation with US-guided optical imaging reconstruction. Radiology. 2005;237(1):57–66.
35. Ntziachristos V, Yodh AG, Schnall MD, Chance B. MRI-guided diffuse optical spectroscopy of malignant and benign breast lesions. Neoplasia. 2002;4(4):347–54.
36. Brooksby B, Jiang S, Dehghani H, Pogue BW, Paulsen KD, Kogel C, Doyley M, Weaver JB, Poplack SP. Magnetic resonance-guided near-infrared tomography of the breast. Rev Sci Instrum. 2004;75(12):5262–70.
37. O'Sullivan TD, Leproux A, Chen JH, Bahri S, Matlock A, Roblyer D, McLaren CE, Chen WP, Cerussi AE, Su MY, Tromberg BJ. Optical imaging correlates with magnetic resonance imaging breast density and reveals composition changes during neoadjuvant chemotherapy. Breast Cancer Res. 2013;15(1):R14.
38. Laughney AM, Krishnaswamy V, Rice TB, Cuccia DJ, Barth RJ, Tromberg BJ, Paulsen KD, Pogue BW, Wells WA. System analysis of spatial frequency domain imaging for quantitative mapping of surgically resected breast tissues. J Biomed Opt. 2013;18(3):036012.
39. Dhar S, Lo JY, Palmer GM, Brooke MA, Nichols BS, Yu B, Ramanujam N, Jokerst NM. A diffuse reflectance spectral imaging system for tumor margin assessment using custom annular photodiode arrays. Biomedical optics express. 2012;3(12):3211–22.
40. Jacobs L. Positive margins: the challenge continues for breast surgeons. Ann Surg Oncol. 2008;15(5):1271–2.
41. Patel R, Khan A, Wirth D, Kamionek M, Kandil D, Quinlan R, Yaroslavsky AN. Multimodal optical imaging for detecting breast cancer. J Biomed Optics. 2012;17(6):066008.

42. Patel R, Khan A, Wirth D, Kamionek M, Kandil D, Quinlan R, Yaroslavsky AN. Delineating breast ductal carcinoma using combined dye-enhanced wide-field polarization imaging and optical coherence tomography. Biophoton. 2013;6(9):679–86.
43. Patel R, Khan A, Quinlan R, Yaroslavsky AN. Polarization sensitive multimodal imaging for detecting breast cancer. Cancer Res. 2014;74(17):4685–93.
44. Azu M, Abrahamse P, Katz SJ, Jagsi R, Morrow M. What is an adequate margin for breast-conserving surgery? Surgeon attitudes and correlates. Ann Surg Oncol. 2010;17:558–63.
45. Backman V, Wallace MB, Perelman LT, Arendt JT, Gurjar R, Müller MG, Zhang Q, Zonios G, Kline E, McGilligan JA, Shapshay S, Valdez T, Badizadegan K, Crawford JM, Fitzmaurice M, Kabani S, Levin HS, Seiler M, Dasari RR, Itzkan I, Van Dam J, Feld MS. Nature. 2000;406(6791):35–6.
46. Brown JQ, Vishwanath K, Palmer GM, Ramanujam N. Advances in quantitative UV-visible spectroscopy for clinical and pre-clinical application in cancer. Curr Opin Biotechnol. 2009;20(1):119–31.
47. Brown JQ, Bydlon TM, Kennedy SA, Caldwell ML, Gallagher JE, Junker M, Wilke LG, Barry WT, Geradts J, Ramanujam N. Optical spectral surveillance of breast tissue landscapes for detection of residual disease in breast tumor margins. PLoS ONE. 2013;8(7):e69906.
48. Fitzgerald AJ, Wallace VP, Jimenez-Linan M, Bobrow L, Pye RJ, Purushotham AD, Arnone DD. Terahertz pulsed imaging of human breast tumors. Radiology. 2006;239(2):533–40.
49. Fitzgerald AJ, Pinder S, Purushotham AD, O'Kelly P, Ashworth PC, Wallace VP. Classification of terahertz-pulsed imaging data from excised breast tissue. J Biomed Opt. 2012;17(1):016005.
50. Fitzgerald AJ, Pickwell-MacPherson E, Wallace VP. Use of finite difference time domain simulations and debye theory for modelling the terahertz reflection response of normal and tumour breast tissue. PLoS ONE. 2014;9(7):e99291.
51. Yngvesson SK, St Peter B, Siqueira P, Kelly P, Glick S, Karellas A, Khan A. Feasibility demonstration of frequency domain terahertz imaging in breast cancer margin determination. In: Proceedings of society of photo-optical instrumentation engineers 2012. Optical interactions with tissue and cells XXIII; vol. 8221, 82210N.
52. St Peter B, Yngvesson S, Siqueira P, Kelly P, Khan A, Glick S, Karellas A. Development and testing of a single frequency terahertz imaging system for breast cancer detection. IEEE J Biomed Health Inform. 2013;17(4):785–797.
53. Ajito K, Ueno Y. Thz chemical imaging for biological applications. IEEE Trans Thz Sci Techn. 2011;1(1):293–300.
54. Ashworth PC, Pickwell-MacPherson E, Provenzano E, Pinder SE, Purushotham AD, Pepper M, Wallace VP. Terahertz pulsed spectroscopy of freshly excised human breast cancer. Opt Express. 2009;17(15):12444–54.
55. Savastru D, Chang EW, Miclos S, Pitman MB, Patel A, Iftimia N. Detection of breast surgical margins with optical coherence tomography imaging: a concept evaluation study. J Biomed Opt. 2014;19(5):056001.

Chapter 2
Familial Breast Cancer and Genetic Predisposition in Breast Cancer

Vighnesh Walavalkar, Ashraf Khan and Dina Kandil

Introduction

Breast cancer is the most common non-dermatologic malignancy in women and it is estimated that approximately one in nine women will develop breast cancer over their lifetimes. In the United States, more than 200,000 new cases of breast cancer were reported in 2010 and breast cancer was responsible for approximately 40,000 deaths (15 % of all cancer deaths) in the same calendar year [1]. The etiology behind developing breast cancer is multifactorial, with many risk factors including diet, lifestyle, reproductive factors and hormonal status. However, a very important risk factor is a genetic predisposition and a positive family history. A genetic influence on mammary carcinogenesis has long been implicated and it is estimated that approximately 10 % of breast cancer patients are carriers of gene mutations susceptible for the development of breast cancer [2]. Of these genes, perhaps the most extensively studied are breast cancer 1, early onset (BRCA1), breast cancer 2, early onset (BRCA2) and Tumor protein p53 (TP53) genes. These are associated with a high risk of developing breast cancer in carriers and hence they are referred to as high-penetrance genes. It should be noted, however, that among breast cancer patients with a strong family history; only 40 % have cancers that are thought to be caused by the above-mentioned three genes [3]. This suggests that in the remaining 60 % of cases, apart from sporadic breast cancers, other genetic pathways are likely involved.

V. Walavalkar · A. Khan · D. Kandil (✉)
Department of Pathology, University of Massachusetts Medical School, UMassMemorial Medical Center, Three Biotech, One Innovation Drive, Worcester, MA 01605, USA
e-mail: dina.kandil@umassmemorial.org

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Ataxia Telangiectasia Mutated Gene (ATM), CHEK2, BRIP1, PALB2, RAD50, PTEN, CDH1, STK11, etc. are examples of genes that are thought to play important roles in breast cancer pathways. In fact, it has now been shown that these moderate penetrance genes along with many low penetrance single nucleotide polymorphisms (SNPs) [4] interact with one another as well as influence pathways involving BRCA1 and BRCA2. Studies have suggested that these genes are involved in complex genetic pathways, some of which are closely related and ultimately are associated with the development of breast cancer. This chapter gives an overview of some of these genes along with the clinicopathologic features of the cancers associated with them. This will be summarized in Table 2.1. We will also briefly touch upon clinical syndromes associated with breast cancer, genetic testing, preventive strategies and certain aspects of management of familial breast cancer in the United States. A summary of these clinical syndromes are presented in Table 2.2.

Genetics of Breast Cancer

High-Penetrance Genes

Breast Cancer 1, Early Onset (BRCA1)

BRCA1 is a large gene located on the long (q) arm of chromosome 17 at position 21 (17q21). BRCA1 is a tumor suppressor gene, which is expressed in response to genomic instability and is influenced by estrogen. Its main function is related to DNA repair including homologous recombination, nucleotide excision repair, and spindle regulation. It also acts as a gatekeeper of cell-cycle progression mainly through checkpoint control [5]. Recent studies have described complex and innovative mechanisms for the localization of BRCA1 to DNA-breaks, including an emerging ubiquitylation-dependent cascade and an association with BRCA2 and genes in the Fanconi anemia pathway [6]. Thus, BRCA1 acts as a regulator of genome stability and its main function is to respond to various types of DNA damage via a complex interaction with BRCA2 and other genes.

Numerous mutations in BRCA1 have been described. The majority of which are point mutations and small insertions/deletions leading to truncated forms of the BRCA1 protein [7]. Large genomic deletions including whole exon deletions have also been detected using more sophisticated methods such as multiplex ligation-dependent probe amplification (MLPA) [8]. Some mutations appear to be more common in certain ethnic groups (founder mutations). The most commonly described is the c.5266dupC mutation (also known as 5382insC or 185delAG), which is seen in up to 2 % of the Ashkenazi Jewish population. However, recent studies have suggested that this mutation may be prevalent in some other ethnic groups where genetic screening of BRCA1 is not routinely performed [9].

Approximately 1 in 1000 individuals in the female population carries a pathogenic mutation in BRCA1. BRCA1 cancers account for approximately 10 %

Table 2.1 A summary of genes associated with the development of breast cancer

Gene	Chromosomal location	Function	Mode of inheritance	Lifetime risk of developing breast cancer	Other major cancer risk	Clinical syndrome association
TP53	17p13.1	Regulates expression of many genes by anti-proliferative mechanisms inducing cell cycle arrest and apoptosis	Autosomal dominant	85–90 %	Soft tissue (sarcomas), bone (osteosarcoma), CNS tumors (choroid plexus tumors), adrenal gland[a]	Li-Fraumeni syndrome, Li-Fraumeni-like syndrome
ATM	11q22-q23	Upstream regulator of proteins involved in double-stranded DNA repair, including BRCA1, TP53, and CHEK2	Autosomal dominant and recessive	~20 % or less	Lymphoproliferative disorders	Ataxia-telangiectasia
CHEK2	22q12.1	Encodes for a threonine/serine kinase that prevents cell proliferation by phosphorylation of proteins involved in checkpoint control	Autosomal dominant and recessive	~20 % or less	Colorectal, prostate	–
BRIP1	17q22.2	Encodes for a DNA helicase that performs BRCA1-dependent DNA repair and checkpoint control	Autosomal dominant and recessive	~20 % or less	Ovary, cervix	Fanconi anemia
PALB2	16p12.2	Acts as a functional bridging molecule linking the DNA repair functions of BRCA1 and BRCA2	Autosomal dominant and recessive	~20 % or less	Pancreas	Fanconi anemia

(continued)

Table 2.1 (continued)

Gene	Chromosomal location	Function	Mode of inheritance	Lifetime risk of developing breast cancer	Other major cancer risk	Clinical syndrome association
CDH1	16q22.1	Encodes for a cell adhesion molecule called E-cadherin	Autosomal dominant	50–60 %	Stomach	Hereditary diffuse gastric cancer syndrome
PTEN	10q23.3	Down-regulates the phosphatidylinositol-3-kinase (PI3K) signal transduction cascade and acts as a tumor suppressor and growth regulator	Autosomal dominant	25–50 %	Thyroid (except medullary carcinoma), endometrium, colon, rectum	Cowden syndrome
STK11	19p13.3	Encodes for serine threonine kinase	Autosomal dominant	~30 %	Colorectal, gastric, pancreatic, ovary	Peutz-Jeghers syndrome
RAD50	5q31	Part of MRN complex along with MRE11 and NBS1, which facilitates double-strand DNA break repair	Unknown	Unknown	Unknown	Ataxia-telangiectasia-like disorder, Nijmegen breakage syndrome (NBS) and NBS-like disorder

[a]Additional risk in TP53 mutations include: gastrointestinal tract cancers (esophageal, gastric and colorectal), genitourinary cancers (renal, Wilms tumor, endometrial, ovarian, prostate), melanoma, thyroid, and lymphoproliferative disorders

Table 2.2 Clinical syndromes associated with breast cancer

	Gene(s) involved	Clinical manifestations	Cancer prevention strategy	Cancer management
Hereditary breast and ovarian cancer syndrome[a]	BRCA1, BRCA2	Early onset of breast and ovarian cancer. Also high risk for early onset prostate, pancreas, skin (melanoma), gastrointestinal tract cancers	• Genetic counseling and standard genetic testing with full gene sequencing and large genomic alterations analysis • Patient awareness and routine monthly self breast exam from 18 years of age onwards • Biannual clinical breast exam from 25 years of age onward • Annual bilateral mammogram and MRI starting at age 25 • Discuss ovarian cancer screening (transvaginal ultrasound and serum CA125 testing every 6 months) • Discuss risks and benefits of chemoprevention • Risk-reducing mastectomies and salpingoophorectomies • Prostate cancer screening in men after age 40	• Individualized chemotherapeutic regimen with poly(ADP-ribose) polymerase inhibitors ± platinum or other combination therapy • Bilateral mastectomies and salpingoophorectomy
Li-Fraumeni and Li-Fraumeni-like syndrome[a]	TP53	Autosomal dominant cancer predisposition syndrome associated with early onset of breast cancer, choroid plexus carcinomas, adrenocortical carcinoma, soft tissue sarcoma and osteosarcomas. Also high risk for many other visceral malignancies and lymphoproliferative disorders	• Genetic counseling and testing • Patient awareness and routine monthly self breast exam from 18 years of age onwards • Biannual clinical breast exam from 25 years of age (or as early as 18 years) onward • Annual bilateral mammogram and MRI from 25 years of age (or as early as 18 years) onward • Colonoscopy every 2–5 years starting 25 years of age onward • Annual skin and neurologic exam	• Surgical management preferred ± individualized chemotherapeutic regimen • Radiation therapy often used as last option as there is a questionable risk for therapy induced secondary malignancy

(continued)