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Review Article

Development of Imaging and Liquid Biomarker Analysis for Breast Cancer Screening: A Review

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Abstract

Background: Breast cancer screening tests could reduce the mortality rates for breast cancer patients. Screening and detection are the keystone of cancer prevention and may significantly minimize the death rates in breast cancer patients for long-term. In this review, we would like to present a comprehensive summary from recent publications of the current development for breast cancer screening, classification of breast cancer based on pathological diagnosis, as well as development of breast cancer detection.

Methods: The sources of the articles were collected from research published in the PubMed, NCBI databases and manual searches without time restriction based on review of the title, abstract and full review of the articles, using the keywords "breast cancer", "diagnostic", "screening", "imaging", "biomarker" and the combination of these terms. The criteria excluded in selecting references were articles that are not written in English, newspapers, and posters.

Results: Of the 146 articles that were selected, there were 103 articles included. Breast cancer screening consists of imaging and pathological assessment such as invasive biopsies of tumor tissue and measurement of biomarkers. The recent development of breast cancer screening utilizing different models and methods like biomarkers were being reviewed. For imaging methods, there are mammography, digital breast tomosynthesis (3D mammography), magnetic resonance imaging (MRI), and ultrasonography. For pathological assessment, there are primary biomarker analysis for breast cancer (estrogen receptor, progesterone receptor, HER2, KI67 index) and liquid biomarker analysis from blood or saliva samples. Additionally, there are some diagnostic kit models for breast cancer screening that were in use such as NanoString nCounter®, MammaTyper®, CellSearch System™, and AdnaTest BreastCancer™.

Conclusion: Each of these methods has its own limitations. Therefore, the development of breast cancer models should be more sensitive, reliable, approachable and less harmful.

Introduction

Breast cancer is one of the highest prevalent cancers in which the incidence rate is higher in developed countries.[1] There are some risk factors such as age, hormone status, family history, genetic predisposition, environment, lifestyle, and population structure that could alter the prevalence of breast cancer which is different every person in many regions.[2] Furthermore, the molecular patterns in primary and metastases tumors of patients are different.[3] Early breast cancer screening is a keystone of cancer prevention and could reduce the mortality rates.[4] The main problems are the lack of community access such as source availability caused by excessively high costs, lack of knowledge, hours of operation, or distance from the access source.[5] Highly sensitive, rapid, reliable, and accessible early-stage breast cancer diagnostic is important factor to decrease mortality rates and improve breast cancer detection quality by reducing recalls of false positive results and unnecessary biopsies.[6]

Breast cancer clinical examinations consist of imaging diagnosis and pathological assessment such as invasive biopsies of tumor tissue and measurement of biomarkers.[7] Imaging techniques contain mammography and ultrasound detection for the breast tissues as a target. Pathological assessment should be conducted based on biomarker detection techniques like core needle biopsy.[8] This paper reviews the current development for early-stage breast cancer screening, classification of breast cancer based on the patient's pathological diagnosis results, and development of breast cancer detection to overcome emerging problems such as availability, accessibility, and patients with certain condition.

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Methods

Search Strategy

The materials for this study were selected from PubMed, NCBI databases and manual search without time restriction based on review of the title, abstract and full review of the articles. The search keyword included: breast cancer, diagnostic, screening, imaging, biomarker and the combination of these terms. The excluded criteria in selecting references were articles that are not written in English, newspapers, and posters.

By using keywords that have been specified, 146 articles were obtained. Of the 146 articles, 103 articles were selected for evaluating the recent development breast cancer screening using different models and methods.

Results

As there no cure yet for breast cancer, there are prevention methods for women to reduce the risk of breast cancer.[9] The primary prevention method is by utilizing prophylactic surgery or chemoprevention for woman with high risk.[9] The secondary prevention method is by utilizing early detections such as clinical breast examination and screening methods like imaging and biomarker analysis that offer the most effective, practical, and viable methods for women around the world.[9] The main objectives of early breast cancer screening are to enable women to be able to undergo less invasive diagnostics and to identify asymptomatic cancer that leads to ideal outcomes before the breast cancer progresses through both physical breast examinations (e.g., mammographic imaging) and pathological assessment as breast cancer screening modalities.[10] The next assessments required are personal medical history, family history, renal and liver function tests, calcium and alkaline phosphatase levels, another physical examination, determining menopausal status and a full blood count.[8] The pathological results consist of the histological type, breast cancer grade (TNM stage), immunohistochemical (IHC) evaluation of breast cancer biomarker such as estrogen receptor (ER) and progesterone receptor (PgR), and human epidermal growth factor receptor (HER2) gene expression status (Figure 1).[11]

Imaging

In most cases, breast cancer is detected by screening test or through analyzing symptoms that occur on patients (e.g., pain or a palpable mass) and associated with the detection of tumor size, metastasis, and would likely require chemotherapy to reduce morbidity and improve the patient's survival rates.[12] Breast magnetic resonance imaging (MRI) or ultrasonography may be considered as additional screenings for high-risk woman.[10] These imaging techniques (Table 1) have advantages for avoiding unnecessary breast biopsies. The early diagnostic method accuracy was determined by sensitivity and specificity from final assessment that were defined as true positive (TP), true negative (TN), false negative (FN), and false positive (FP). The calculation formula for sensitivity is = (TP/(TP +FN) × 100 and for specificity is = TN/(TN+FP) × 100.[13]

Mammography and Digital Breast Tomosynthesis (3D mammography)

Mammography is the basic method of breast cancer diagnostic that only has average sensitivity and could reduce inpatient with high dense breasts because of overlying breast parenchyma or lesions from overlapping tissues.[4] Mammography also has disadvantages such as the use of ionizing radiation, relatively high false-output rates, and providing uncomfortable examination for patients.[14] The impact of such disadvantages is that the patients must be recalled for another exact assessment and require improvement for them to be viable for breast cancer screening.[15] There are digital breast tomosynthesis (DBT) and contrast-enhanced digital mammography to improve the limitations of the conventional mammography like the specificity.[16] Contrast-enhanced mammography techniques have the potential to encourage good initial results with 85.2% sensitivity and 66.1% specificity.[17] Contrast-enhanced mammography could reduce radiation exposure, health care costs, and false-positive rates.[18] The development of digital mammography creates digital breast tomosynthesis that could provide analysis of

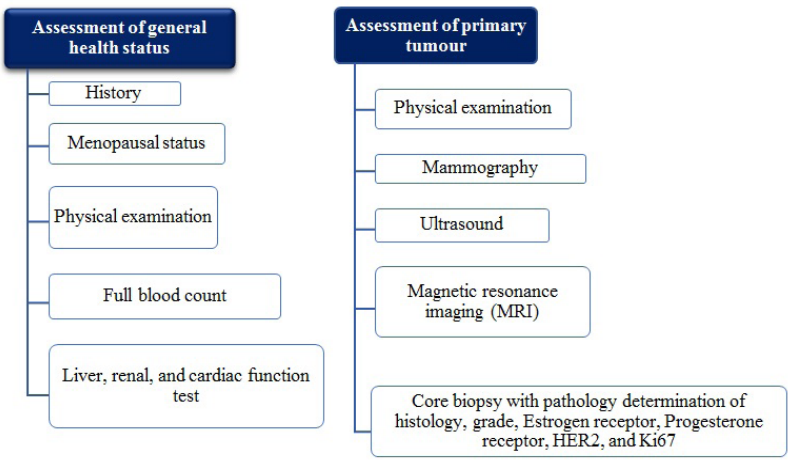

Figure 1. Diagnostic steps for early breast cancer patient.

3D mammographic data that presents high detail and answering some disadvantages from overlapping tumor tissue.[19] Digital mammography may have drawbacks that reduces sensitivity because fibroglandular tissue may be overlying tumors.[10] Digital breast tomosynthesis using X-ray kit that could move over a limited arc angle and reconstruct the tissues in thin slices to reduce overlapping tumor tissue.[16] The addition of digital breast tomosynthesis (DBT) may improve the detection rate of breast cancer and reduce patient recalls rate.[20] One concern on using this method for breast cancer screening is the digital breast tomosynthesis contains double radiation dose over conventional digital mammography alone.[12]

Magnetic Resonance Imaging (MRI) and Ultrasonography

Both magnetic resonance imaging (MRI) and ultrasonography are decent tools for evaluating and diagnosing abnormalities of breast tissues especially for specific populations such as very high-risk women with mutations of BRCA1 and BRCA2 or women with dense breast.[10]

Magnetic resonance imaging (MRI) displays multiple cross-sections image by involving magnetic field. The resolution of magnetic resonance imaging (MRI) could be increased by applying contrast agent[4] and already been recommended for detecting breast cancer in high risk breast cancer patients.[21] Magnetic resonance imaging (MRI) is less specific but more sensitive compared to mammography or ultrasound for detecting small tumors in patients with high-risk.[22] Several studies recommend combination of mammography and magnetic resonance imaging (MRI) for women with high breast cancer risk. The benefit is demonstrated by comparing a group of women with BRCA1 and BRCA2 mutations.[23]

Breast ultrasonography is a good method as it is widely available, cost effective, and could improve examination sensitivity through the detection of the breast cancer mass shape.[24] Some observational and clinical studies have shown that the combination of ultrasonography and mammography could increase the detection rates and sensitivity of breast cancer screening in women which have dense breasts.[25] Breast ultrasonography has already been introduced as an additional screening for high-risk patients.[26] However, there are some disadvantages of breast ultrasonography such as possible failures in screening many tumors due to the similar features of cancerous and normal tissues. Moreover, this method requires experienced radiologists and could affect the specificity and sensitivity of the results.[4]

Biomarker Analysis

There are certain indications of cancer progression called biomarkers (Table 2) that could be expressed in/on tumor tissues such as biomolecules from microRNAs, some mutated genes, and cell surface receptor proteins.[27] As a diagnostic tool, biomarkers assemblies demand such non-

Table 1. Imaging methods for early breast cancer screening.

Type	Use	Sensitivity*	Specificity*	Advantage	Disadvantage	Ref.
Mammography	Mass detection. Displays bone, blood vessel and soft tissue image.	67.8%	75%	First recommendation for early breast cancer screening, proven to reduce breast cancer specific mortality.	Radiation exposure, relatively high false-positive rates and false-negative rates, not suitable for women (patients) with high risk such as dense breasts, and slightly uncomfortable assessment.	4, 12
Ultrasonography	Detects the mass shape of breast cancer at an early stage.	83%	34%	Detecting an early-stage breast cancer in patients with the dense parenchyma	Requires experienced operator during examination and displays image with low resolution.	4, 24
Magnetic Resonance Imaging (MRI)	Displays small details images of soft tissues.	94.4%	26.4%	Screening for patient with high risk especially for young women or patients with dense breasts.	Overestimation of tumor size, expensive method.	4, 12,22
Contrast-enhanced Mammography	Detects area that shows vascularization in patient.	85.2%	66.1%	Improving sensitivity for conventional mammography, better lesion representation in dense breast where the image may be blocked by fibroglandular tissue	Low specificity, limited evidence for breast cancer screening.	17
Digital Breast Tomosynthesis	Examines actual breast lesions, allows better separation in tumorous and normal tissues illustration of lesions.	90.77%	96.49	May improve lesion detection and reduces false positive and recall rates.	Radiation dose approximately is twice that of mammography.	6, 19, 20

* Breast composition and the types of cancer could affect sensitivity and specificity of the methods.

invasive techniques and should be differently identified in healthy individuals.[28] The serum could be collected as common analytes for a biomarker as expressed on the cell surface extra cellular domains (ECD).[27] Breast cancer biomarkers have two classifications: overexpressed biomolecules-based biomarkers and stage-dependent biomarkers.[29] Biomarkers may contain genetic sequencing information for some individuals that have BRCA mutation.[7] Basic expression techniques for biomarkers are enzyme linked immunosorbent assay (ELISA), immunohistochemistry (IHC), and radioimmunoassay.[30] Core biopsy with following immunohistochemical breast cancer molecular subtypes evaluation is currently the basic method for breast tissue assessment as it has relatively high sensitivity compared to other methods.[31] Immunohistochemical examination is utilized to facilitate the classification of breast cancer subtypes.[32] The main molecular biomarkers that is related to breast cancer are progesterone receptor (PR), estrogen receptor (ER), Mib1/Ki-67 proliferation index and human epidermal growth factor receptor (HER2) as they are remarkably established in the standard care of breast cancer patients.[33] ER and PR could stimulate the growth of breast epithelium, play an important role as sex steroid receptors and could express around 75% of all breast cancers. Additionally, poor prognosis could also be related to overexpression of HER2.[34] (Table 2).

Estrogen Receptor (ER) and Progesterone Receptor (PR)

Sexual hormones usually provide an impact to growth of breast cancer tissue. Estrogen receptor is one of the most notable biomarkers because estrogen receptor acts as transcription factors that promote survival, proliferation, and invasion of the cell.[33] Estrogen receptor is ligand-regulated. The DNA-binding domains are the main components of estrogen receptor which specifically binds with high affinity on estrogen response elements (ERE sequence) and organizes the transcription rates of ligand-binding domain.[35] There are ERα and ERβ which are the two forms of estrogen receptor that are differentially expressed in tissues. Both ERα and ERβ manage cell differentiation and proliferation by binding estradiol in the normal mammary gland.[36] ER-positive patient could reduce recurrence and mortality from breast cancer by using ER as the target therapy and endocrine therapy (tamoxifen and aromatase inhibitors) as the treatment.[32] Nowadays, immunohistochemistry is the standard practice evaluation of estrogen-progesterone receptor expression.[37] There are guidelines that establish the inspection criteria and proficiency testing for hormone receptor to increase its accuracy. The specimens of breast resection must be arranged as quickly as possible (within 1 hour from resection) in a fixative with adequate volume.[38]

Human Epidermal Growth Factor Receptor 2 (HER2)

The human epidermal growth factor receptor 2 (HER2) genes are localized on chromosome 17 and they are regularly expressed at low levels in all epithelial cells. HER2 are one of the significant components for cancer survival and proliferation.[33] High levels expression of mRNA and protein product by HER2 genes amplification could conduct self-sufficiency and oncogenic resultant signaling in growth signals, continuous angiogenesis, uncontrolled growth, and amplify metastasis processes that could encourage carcinogenesis.[32] The total results of HER2 that are amplified in patients range approximately from 15–30% of breast cancers cases.[39]

Immunohistochemistry for testing HER2 protein overexpression has been developed, and may become the standard procedure for detecting invasive breast carcinomas.[40] The amplification of HER2 gene could be analyzed by fluorescence (FISH), silver-enhanced (SISH) or chromogenic in situ hybridization (CISH) and directly linked to mRNA and protein expression levels that could be analyzed through ELISA test, Western blot, immunohistochemistry, real time PCR or Northern blot.[33] Immunohistochemistry has already been assessed as the standard test in determining the HER2 status, which has advantages such as quicker results, the ability to display morphological tumor appearance, and the ability to maintain stained preserved tissues to degrade slower over time.[32]

MiB1 / Ki67

Mib1/Ki-67 is a biomarker measured through proliferation index as the parameters for predictive and prognostic markers. In most cases, breast cancer patients experience worse outcomes as they are expressing high levels of Ki67.[32] Mib1/Ki-67 index decreases for patients who are provided with post-treatment of neoadjuvant therapies. It becomes a decent predictor for improved clinical outcomes. However, the ASCO guidelines have not included Mib1/Ki67 index as a primary assessed marker for breast cancer prognosis due to the lack of standardization of testing and the interpretation of this index.[33]

BRCA

The BRCA genes comprehend a group of tumor suppressor genes.[41] Patients with BRCA mutation carriers could increase their lifetime risk of breast cancer.[42] In reference to the previous studies, there are 70% cumulative risks for BRCA carriers (BRCA1 & BRCA2) who are diagnosed with primary breast cancer. BRCA-related tumors frequently show different histopathological features that are incompetently differentiated but also highly proliferative.[43] Partial BRCA1 protein could be produced by a mutation in exon 11 that is encoded by the known exon 11 splice variant and it features a different function from whole BRCA1 protein.[42]

Liquid Biopsy Biomarker

There are several studies that initiates on the capability of liquid biopsy to confirm the genomic profile, monitor responses of therapy, and evaluate the emergence of

Table 2. Primary protein biomarkers for breast cancer.

Protein	Description and Function
Estrogen and progesterone receptor (hormone receptor)	Estrogen and Progesterone Receptor may encourage cell proliferation, tumor invasion and survival. Estrogen plays an important role for translocation by binding to its receptor into the nucleus as a transduction signal and as a transcription factor for several physiological responses in many target organs.
HER2 (human epidermal growth factor Receptor)	High level expressions of mRNA and protein product by HER2 genes amplification could conduct self-sufficient and oncogenic resultant signaling in growth signals, continuous angiogenesis, high proliferation rates, amplify metastasis processes and invasion that could encourage carcinogenesis.
MIB1/Ki-67	Mib1/Ki-67 is a biomarker that is measured by proliferation index as parameters for predictive and prognostic marker. Although it is widely used, the ASCO guidelines have not included Mib1/Ki67 index as a primary assessed marker for breast cancer prognosis due to the lack of standardization.

resistance from patients.[44] In addition from the blood, there are several other body fluids like urine,[45] saliva,[46] cerebrospinal fluid,[47] pleural effusions,[48] and stool.[49] Serum or plasma that are utilized as biomarkers samples are potential for breast cancer screening as they accommodate valuable cellular and molecular content in the blood, which provide data about individual health information and could develop a great noninvasive diagnostics for breast cancer.[50]

Some protein and peptide profiling in biological fluids has already become an interesting novel biomarkers for cancer patients.[51] This method utilizes mass spectrometry (MS) as a tool to differentiate proteomic scheme of healthy individuals as controls and cancer patients.[52] The early-stage of breast cancer detection emerged due to the increase of abnormality of total biomarkers from breast cancer patients that were up or down-regulated in comparison with healthy controls population.[53]

Blood-based diagnostic assay

There are blood-borne tumor biomarkers have been introduced as a diagnostic assay to evaluate malignancy prior to the clinical diagnosis, such as the human epidermal growth factor receptor (HER2), carcinoembryonic antigen (CEA),[54] the oncogenic protein RS/DJ-1, and circulating cytokeratin fragments.[55]

Blood-based Test using Multiple Reaction Monitoring

There is a blood-based test utilizing multiple reaction monitoring (MRM) as the method and measured by mass spectrometry that quantifies as 3 peptides: apolipoprotein C-1 (APOC1), carbonic anhydrase 1 (CAH1), and neural cell adhesion molecule L1-like protein (NCHL1) that illustrate different concentration level between healthy individuals as a control and breast cancer patients.[56] APOC1 plays a vital role in lipoprotein metabolism that binds to fatty acids[57] and reduces the addition of estrogen in cells. The amount is 0.7 times less in stage 1 breast cancer patients than the condition of healthy women.[58] CAH1 enzymes are overexpressed and they increase rapidly through angiogenesis which is a key mechanism for tumors to develop to cancer.[59] The amount is 1.61 times higher for stage 1 breast cancer patients than the condition of healthy women.[58] NCHL1 is closely related to cancer expression and metastasis[60] and the amount is 1.4 higher more for stage 1 breast cancer patients than the condition of healthy women.[58]

Blood-based Test for Detecting Copper (Cu)

The expression level of ATOX1 for patients may become a breast cancer biomarker for early stages diagnostics. Antioxidant 1 copper chaperone (ATOX1) plays an important role in cell migration process which is a core phase in metastasis.[61] Copper (Cu) is one of the constituents of many enzymes that is required for several mechanisms in cancer such as angiogenesis, metastasis and proliferative immortality.[62,63] The Cu concentration levels have been increased for breast cancers patients that have already developed a distant metastasis.[6]

Blood-based Test for Biomarker Fragments (ctDNA, CTCs, EXOs, MiRNA)

Novel approaches on the development of breast cancer diagnosis must provide potential biomarkers that contain relevant clinical information, meets the requirement, and less invasive methods (Figure 2). There are fragments in liquid biopsy samples such as circulating tumor DNA (ctDNA), circulating tumor cells (CTCs) or exosomes (EXOs) that are detached from tumorous cells (necrotic or apoptotic).[44] Low concentration of cell free DNA detected in the healthy person's blood and the quantity could be increased in breast cancer patient.[64]

Cell free DNA (cfDNA) indicates the total DNA fragments in the blood samples that develop from three different sources which actively produce DNA: necrotic cells, apoptotic cells, and viable cells.[65] The main purpose of cfDNA-based analysis is to gather information towards the genetic changes in DNA fragments that are obtained from cancer cells such as circulating tumor DNA (ctDNA),[66] which could be analyzed by sequencing or digital polymerase chain reaction (dPCR).[67] There are some disadvantages from analyzing ctDNA which are the fraction of ctDNA is relatively low from the total cfDNA in cancer patients,[68] only applicable for minority patients, as well as more expensive.[69] The amount fractions of ctDNA are ranging from 0 (undetectable) to 11.7%. There is no economical way to evaluate the amount of ctDNA fraction within the total of cfDNA[66] and still requires evidence in clinical trial.[70]

There are tumor cells called circulating tumor cells

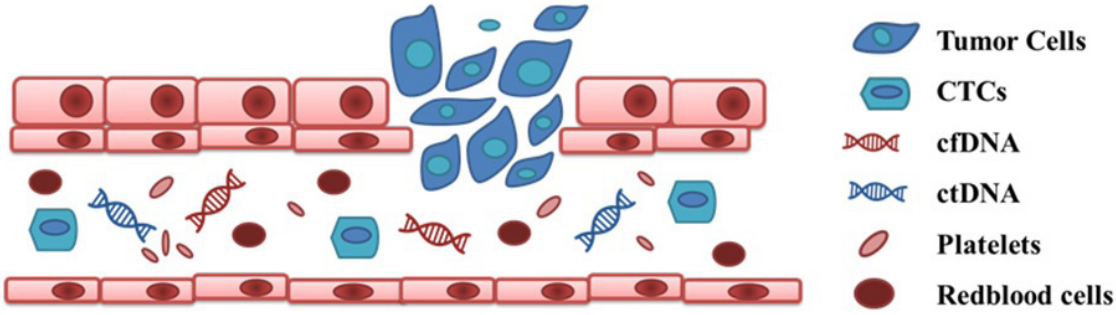

Figure 2. Biomarker fragments of breast cancer patient in blood vessel.

(CTCs) that possibly have been passively released from the major tumor and metastatic lesions into the bloodstream.[44] CTCs could be measured from blood samples of cancer patient and the total CTCs are correlated to overall survival and treatment outcomes.[71] The concentration levels of CTCs that were detected in blood samples are relatively low, however it varies on the tumor types.[72] CTC detection methods usually consist of several steps such as primary enrichment (due to low concentration), and cell isolation from blood cells with epithelial markers.[73] There are two common kits for detecting CTCs, CellSearch CTC test and AdnaTest.[74]

Blood cells, smooth-muscle cells, platelets, endothelial cells, and immunocytes are known to be able to release exosomes which have significant roles in switching molecular information among cells.[75] They have already indicated that they contain proteins as well as some nucleic acids such as deoxyribonucleic acid (DNA), messenger RNA (mRNAs), and micro RNA (miRNAs). It is also shown that they can arrange the action of recipient cells. Exosomes could possibly be utilized as biomarkers of cancer.[44] Furthermore, exosomal miRNAs could be handful in cancer development as they could encourage angiogenesis and stimulate metastasis.[76]

MicroRNAs are endogenous RNA molecules that contain between 19–25 nucleotides and have essential roles in post-transcriptional level as gene regulatory.[77] MicroRNAs could develop combination with protein-coding genes messenger RNAs (mRNAs) and would lead to mRNA translational degradation or repression.[78] MicroRNAs have a vital role in many cellular processes like differentiation, apoptosis, and proliferation. MicroRNAs alteration could also bring harmful transformation.[79] Several circulating miRNAs are diversely produced in the serum or plasma and could become potential biomarkers for breast cancer as the amount is different for healthy individuals and breast cancer patients. According to several studies, the most consistently upregulated miRNA is miR-21 as it functions as oncogene, while the most downregulated is miR-145 as it functions as tumor suppressor.[80]

Salivary-based diagnostic assay

Biomarker research is constantly developing to the point where saliva is introduced as a very good diagnostic sample through technological advancements that could be collected non-invasively, simple, and could be gathered regularly without bringing discomfort to the subject.[81] Salivary biomarkers analysis provide additional advantages such as monitoring clinical condition status and predicting diseases,[82] but it requires combinatorial analysis of the biomarker profile to achieve appropriate level of specificity and sensitivity.[28]

Vascular endothelial growth factor (VEGF), Carcinoembryonic antigen (CEA), and Epidermal growth factor (EGF)

Breast cancer metastasis and tumor growth could be elevated by angiogenesis mechanism and some angiogenic components such as vascular endothelial growth factor (VEGF), carcinoembryonic antigen (CEA), and epidermal growth factor (EGF). They detected through enzyme-linked immunosorbent assay (ELISA) and could be found in saliva samples of breast cancer patients.[83] The level of those biomarkers are increased in the saliva samples of breast cancer patients in comparison with healthy individuals, mostly when those biomarkers were analyzed together as a combination.[28]

Autoantibodies - Mucin1 (MUC1), Human Epidermal Growth Factor Receptor (HER2)

There is a high interest of exploration in autoantibodies against tumor biomarkers that could be evaluated in saliva.[84] Autoantibodies that are expressed against tumor biomarkers could offer a beneficial approach such as providing noninvasive method for breast cancer diagnostics.[28] Mucin1 (MUC1) is a transmembrane glycoprotein that is overexpressed by around 90% in breast tumor and it performs a crucial part in development of the cancer. When MUC1 is overexpressed, it would stimulate growth of cells, resistance of therapy, and metastasis in cancer.[85] Human epidermal growth factor receptor (HER2) is one of the biomarkers that is already detected and overexpressed in breast cancer patient, so it could induce cell migration and potentially become a metastatic factor.[86] Autoantibodies against MUC1 and HER2 have already been investigated by using immunoglobulins (IgM and IgG) and has been detected by enzyme-linked immunosorbent assay (ELISA) test. The immunoglobulins were remarkably higher in breast cancer patients than in healthy individuals.[87]

Sialic acid

Sialic acid is biologically notable for glycoconjugates and could be altered in cancer patients, thus the sialylation processes of cell surface glycoconjugates are increased and could cause malignant cancer progression.[88] Sialic acid concentration of salivary samples in breast cancer patients have been significantly increased compared to healthy individuals.[89] From the result, it could be concluded that sialic acid establishes clinical importance as a diagnostic marker.[28]

Metabolites – Proline, Valine

Metabolites are one of the biomarker classes that is widely discovered in saliva samples of breast cancer patients utilized for diagnostic purposes.[28] Some studies showed significant changes in amino acid (basic metabolites) profile of breast cancer patients such as proline and valine. The analytical techniques that are utilized for detecting metabolites are Gas chromatography–Mass spectrometry (GC-MS), Liquid chromatography–Mass spectrometry (LC-MS), and Nuclear magnetic resonance (NMR spectroscopy).[90]

Problem and Risk for Breast Cancer Screening

Although breast cancer screening has several benefits for women, it also hosts potential harms such as the side effect of screening. Balancing between the advantages and the harms of breast cancer screening may be rather complicated due to several considerations such as establishing harms possibilities, deciding the prime ages to start regular screening, determining the best intervals of screening test, using the relevant multiple imaging methods, and preferences of women concerning to screening.[91] Breast cancer screening illustrated some risks such as false-positive diagnostic results,[92] anxiety,[93] radiation exposure,[94] pain during procedures,[95] and overdiagnosis.[96]

Diagnostic Kit Models for Breast Cancer Screening

Reliable diagnostic test is essential for breast cancer classification and biomarkers such as human epidermal growth factor receptor 2 (HER2), estrogen receptor (ER), and progesterone receptor (PR).[97] The best analysis for the molecular research should be specific, reliable, sensitive, and easy to perform.[98] There are some diagnostic kit models that have already been used for breast cancer screening such as NanoString nCounter®, MammaTyper®, CellSearch System™, and AdnaTest BreastCancer™.

NanoString nCounter®

NanoString nCounter® gene expression system is a digital quantification technology based on RNA that performs color-coded multiplexed target molecule. It establishes the transcripts counts of mRNA from a limited quantity of total RNA without any amplification.[99] NanoString nCounter® gene expression system was performed for quantizing mRNA expression level of human epidermal growth factor receptor 2 (HER2), estrogen receptor (ER), and progesterone receptor (PR).[98]

MammaTyper®

MammaTyper® is a diagnostic test that performs quantification for messenger RNA (mRNA) expression of biomarker genes such as PGR (PR), MKI67 (Ki-67), ERBB2 (HER2), and ESR1 (ER) by real-time quantitative polymerase chain reaction (RT-qPCR). MammaTyper® also classifies the results into different molecular subtypes i.e. HER2 positive (non-luminal), triple negative (ductal), Luminal A-like, and Luminal B-like (HER2 positive/HER2 negative).[100]

CellSearch System™ and AdnaTest BreastCancer™

Both CellSearch System™ and AdnaTest BreastCancer™ are diagnostic tests for detecting circulating tumor cells (CTCs) in blood samples of breast cancer patients.[101] Those methods contain the cell-enrichment step and the detection step. Mainly, the cell-enrichment step requires antibody-based magnetic capture towards to the epithelial cell adhesion molecule-1 (EpCAM) as a target,[102] and could be detected using immunofluorescence for CellSearch System™ and by measuring tumor-associated transcript (MUC-1, HER2, and GA733-2) with reverse transcriptase-polymerase chain reaction (RT-PCR) for AdnaTest™ BreatsCancer.[103]

Conclusion

Early-stage detection of breast cancer may significantly minimize death rates in breast cancer patients for long-term. The realization of early diagnostics and screening programs are fundamental principles of cancer prevention. This paper summarized the screening methods and kind of biomarkers which are frequently available for diagnosing early-stage breast cancer. The recent development of breast cancer screening that utilizes different models and methods such as biomarkers were being reviewed. The development of breast cancer models should be more sensitive, reliable, approachable and less harmful.

Author Contributions

AS: Drafting the work, TR: The conception and design of the work and revising the work, RA: The conception and design of the work, and revising the work. All the authors agreed to the published version of the manuscript.

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Conflict of Interest

All authors here claim no involvement in a conflict of interest, financial or otherwise.

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