ERURJ **2024**, *3*, 4, 1659-1679
(Review)

Progress of Breast Cancer Biomarkers for Optimized Management: Are We There Yet?

Sameh H Mohamed [1, *], Ahmed M. Reda [1], Ishak E. Kerilos [2]

[1] *Department of Biochemistry, Faculty of Pharmacy, Egyptian Russian University, Cairo 11829, Egypt.*

[2] *Department of Pharmaceutics and Pharmaceutical Technology, Faculty of Pharmacy, Egyptian Russian University, Cairo 11829, Egypt.*

*Corresponding author: E-mail address: Sameh-hosam@eru.edu.eg**,** Tel.#: +2-02-2860-9114

Received 17th May 2024, Revised 21st August 2024, Accepted 22nd September 2024.
DOI:10.21608/erurj.2024.290352.1143

ABSTRACT

Breast cancer continues to be a significant global health burden, necessitating constant advancements in biomarker discovery and management strategies. This review article aims to provide a comprehensive overview of the progress made in breast cancer biomarkers and their clinical implications, as well as the current state of breast cancer management. We begin by highlighting the importance of biomarkers in breast cancer diagnosis, prognosis, and prediction of treatment response. Various molecular markers, including hormone receptors (estrogen receptor and progesterone receptor), human epidermal growth factor receptor 2 (HER2), and Ki67 are mentioned. Moreover, we explore emerging biomarkers in breast cancer, including circulating tumor cells, cell-free DNA, and particularly, breast cancer stem cells, which show promising potential for improved diagnostics, personalized treatment selection, and monitoring of disease progression. Accordingly, we address the importance of multidisciplinary collaboration and the integration of biomarkers into clinical decision-making processes. In conclusion, this review article provides a comprehensive assessment of the progress in breast cancer biomarkers. By examining the current state of biomarker discovery for optimized clinical application and treatment modalities, we aim to shed light on the advancements achieved thus far and identify the remaining gaps in order to pave the way for improved breast cancer care and patient outcomes.

Keywords: Breast cancer; biomarkers; breast cancer stem cells; hormone receptor; personalized treatment

1. Introduction

Breast cancer is a significant health issue because it is the primary cause of cancer-related mortality among women. Over the past ten years, significant progress has been made in the management of people with breast cancer, mostly as a result of earlier identification and better treatments. Heterogeneity in breast cancer, however, continues to be a significant problem that influences patient outcomes. The categorization of breast tumors based on histology and molecular parameters has been crucial in shifting clinical practice away from a "one size fits all" approach to one that is more individualized. Initially, breast tumors were categorized based on gene expression, tumor features, and clinical results [1]. The Breast Cancer Consensus Subtype (BCCS) employs the entire transcriptome to classify tumors into subtypes with a predetermined prognosis, whereas another expression signature, PAM50, uses the expression level of 50 genes [2]. Despite the identification of over ten subtypes of breast cancer, only four were clinically significant: HER2-enriched (Human epidermal growth factor receptor 2), TNBC (Triple-Negative Breast Cancer), luminal A, and B [3]. Estrogen receptors (ER) and progesterone receptors (PR) are expressed in luminal A and B tumors, and the KI-67 proliferative marker is used to determine the degree of proliferation in each tumor. The HER2 oncogene is amplified in HER2-enriched tumors. The most aggressive subtype that lacks HER2, PR, or ER expression is TNBC. This classification allows for the treatment of tumors expressing PR and ER with hormone therapy, while tumors overexpressing the HER2 receptor can be treated with anti-HER2 targeted therapy. Conventional treatment with a combination of anti-mitotic and Deoxyribonucleic acid (DNA)-targeting drugs is advised for TNBC. The emergence of drug resistance and subsequent relapse is a consequence of breast cancer and a leading cause of cancer-related mortality, despite the fact that 87% of patients benefit from these treatments. Individuals who appear to have comparable tumor types do not react to the same medications in the same way [3]. Thus, for effective therapy, patient classification based on drug response prediction is critically needed. In addition to traditional treatments including chemotherapy, targeted therapy, and endocrine therapy, other treatments have also been investigated. These consist of immunotherapy, tyrosine kinase inhibitors (TKIs), clustered regularly interspaced short palindromic repeats (CRISPR), cyclin-dependent kinase 4/6 (CDK 4/6)

inhibitors, medication repurposing, electrochemotherapy, and nanotechnology methods [4] as summarized in Figure 1.

When it comes to the preoperative work-up, diagnosis, and screening of breast cancer, breast imaging is essential. Patients with early-stage breast cancer are not advised to screen for recurrence outside of normal mammography due to the high cost and little chance of positive results. Recurrent diagnostic imaging, however, is standard for patients with metastatic breast cancer (MBC), up to every six weeks. Particularly in clinical trials, the Response Evaluation Criteria in Solid Tumors (RECIST 1.1), which uses imaging to characterize lesions, has gained widespread acceptance as a standard method of measuring tumor response to therapy [5]. However, the use of gadolinium or iodine-based contrast can be hazardous, and imaging is costly and time-consuming. Furthermore, before further imaging is obtained, the disease may advance while receiving an ineffective treatment for a considerable amount of time. Biomarkers can offer more information about a patient's prognosis and reaction to therapy. HER2, PR, and ER tissue expression are among the many biomarkers that are now employed in the treatment of breast cancer. The ability of serum biomarkers to identify therapy response or disease progression before imaging makes them particularly interesting. Serum indicators, however, are less well-established in the treatment of breast cancer [5].

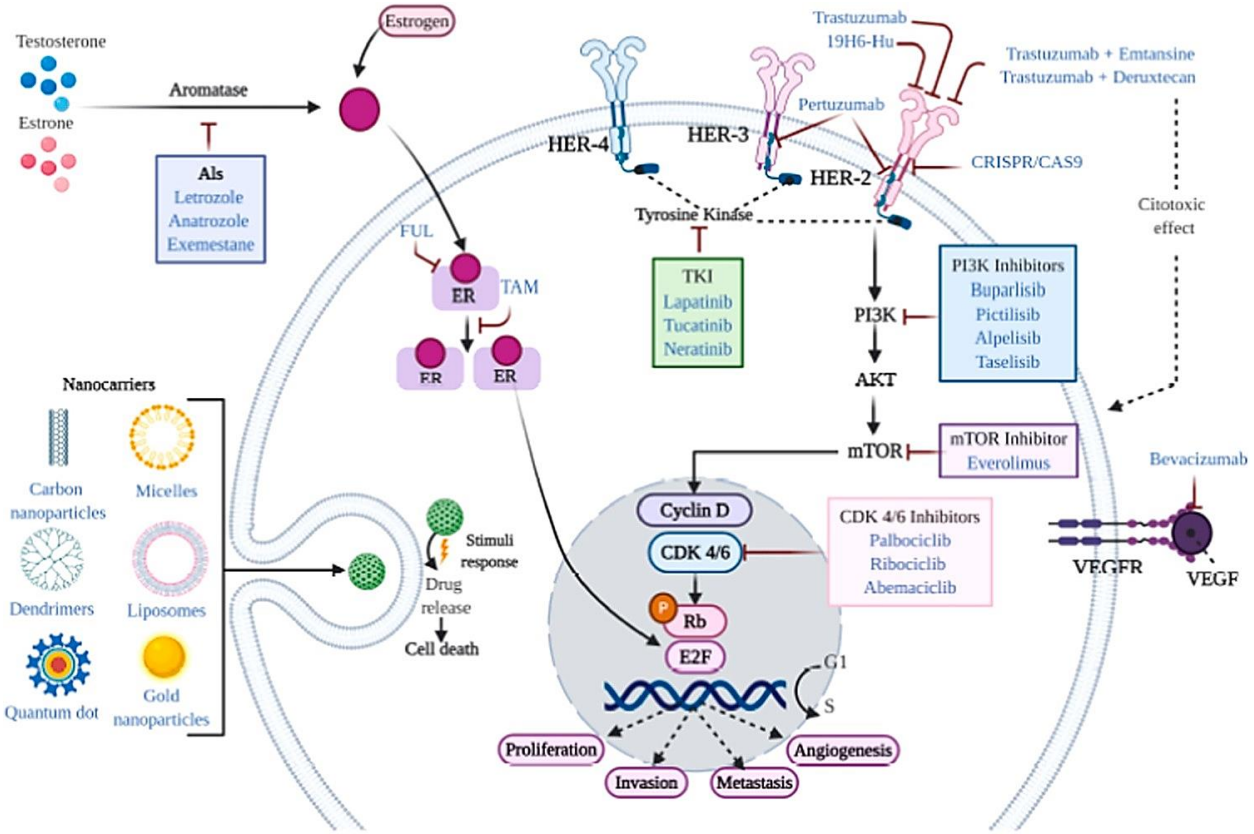

Fig. 1. Treatment options for breast cancer, both traditional and novel, that address the disease's resistance [4].

Finding reliable and accurate biomarkers has the potential to significantly improve clinical practice in a number of ways, including decreased toxicities and financial savings from avoiding the use of ineffective treatments, as well as better outcomes from the selection of the right treatment for suitable patient subpopulations. Companion biomarkers are interesting tools that help clinicians choose the best indication for each of the many novel medicines being developed to treat breast cancer. Thus, in order to optimize clinical application and treatment modalities of breast cancer, we present a thorough overview of the advancements made in breast cancer biomarkers, types, and classifications in this review, along with their clinical implications.

2. Biomarkers in Breast Cancer: Current status

Prognosis and treatment have long been determined by ER and PR expression as well as the HER2 (gene name ERBB2) amplification status. The genetic foundations of invasive breast cancer (IBC) have been discovered more recently, which has led to new developments in

biomarker testing. These tests include tissue or circulating tumor DNA sequencing, immunohistochemistry (IHC) stains to predict therapeutic response, and multi-gene assays (MGAs) that use gene expression analysis to produce prognostic and predictive scores. Furthermore, assays that forecast the outcome of "tumor-agnostic" treatments which focus on tumor-specific genetic changes or variations in protein expression are now available, independent of the type of tumor [6]. Although patients benefit from the growing range of therapy alternatives, pathologists find it difficult to keep up with the quick advancements.

Pathologists are being requested more and more to choose the right molecular assays or to automatically order these procedures. To be legitimate, the test must meet the specific indications and patient/tumor circumstances for each assay. The pathologist must also select the appropriate tissue and block before beginning any examination. This can become more difficult when there are several primaries, metastases, recurrences, a history of neoadjuvant therapy, etc., especially in cases of breast cancer [6].

To summarize, predictive and prognostic biomarker testing in breast cancer included ER and PR IHC, PIK3CA testing, multi-gene assays, immune checkpoint inhibitors, Ki67 IHC, HER2 testing and next-generation sequencing (NGS) assays [6]. In the following sections, a brief description of the most renowned and recognized biomarkers for breast cancer is given below, along with the new trends and directions towards the highest accuracy and optimized personalized medicine for breast cancer.

3. Predictive and Prognostic Biomarkers

Treatment selection or identifying patients most likely to benefit from a specific therapy, is done with the use of predictive biomarkers. These biomarkers have the ability to predict tumor resistance as well as the effectiveness of a particular treatment. One well-known phenomenon that indicates an illness that is resistant to pharmaceutical therapy is drug resistance. Resistance may be adaptive, meaning it develops as a result of the therapy, innate, meaning it exists before the treatment even begins, or acquired (Figure 2) [3,7].

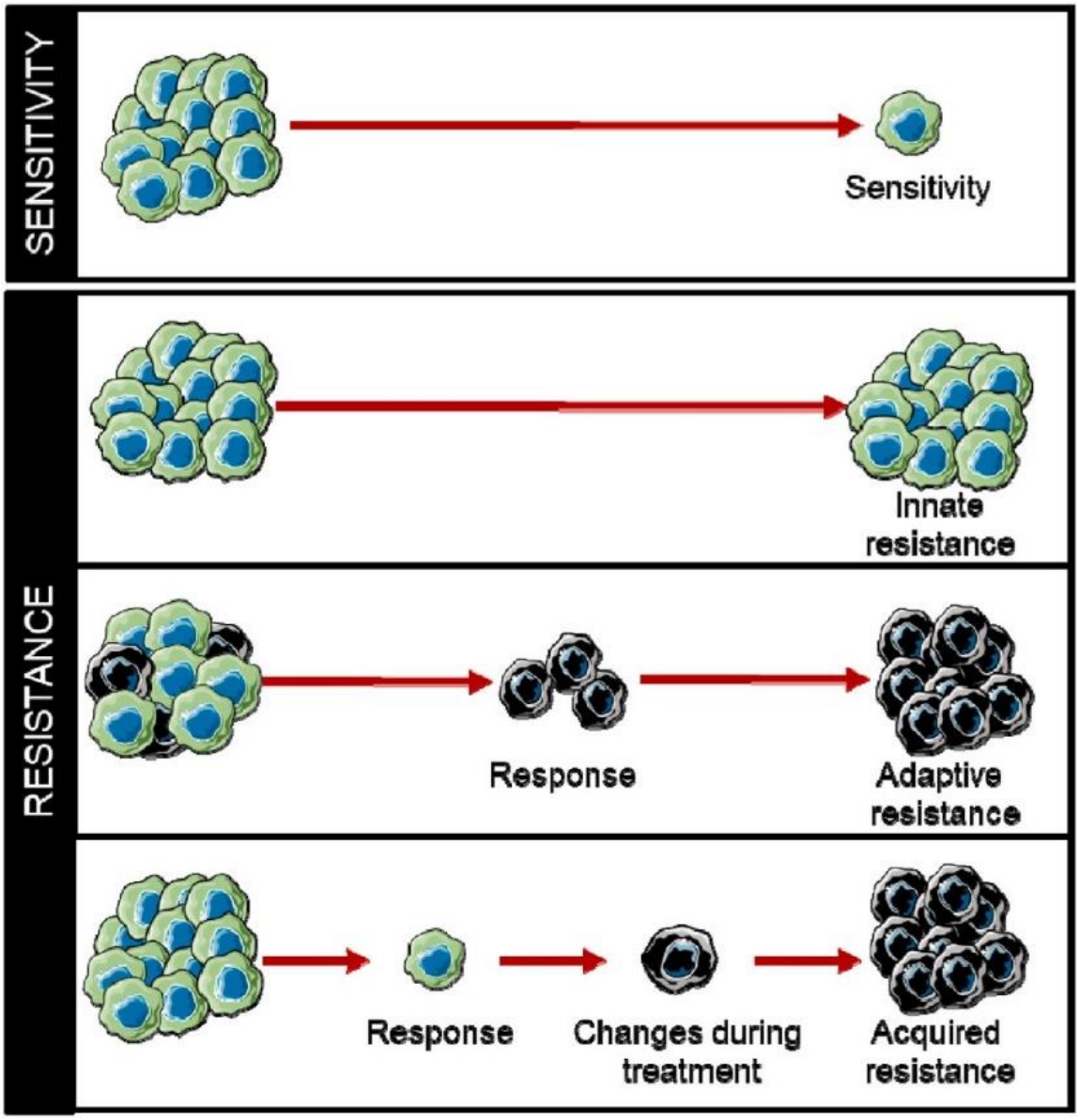

Fig. 2. An illustration of a tumor's sensitivity and resistance to therapy. Tumor contraction serves as a symbol for sensitivity. Innate resistance biomarkers are present in tumors at diagnosis and indicate that treatment will not slow the tumor's growth. When a treatment effectively destroys sensitive tumor cells but spares previously resistant ones, adaptive resistance develops. Tumor relapse occurs when the residual tumor cells proliferate and adapt. Tumor cells change after being exposed to treatment, which leads to acquired resistance [3].

It is possible to measure two types of predictive biomarkers: biomarkers of resistance and biomarkers of response, often known as biomarkers for therapy benefit. The likelihood of a successful outcome and the response rate are both correlated with the expression of response

biomarkers. These biomarkers make it possible to identify patients who are more likely to react favorably to a particular treatment. Conversely, therapeutic resistance biomarkers assist in identifying patients who should not receive treatment, hence avoiding needless medical intervention [8].

Finding and validating a predictive biomarker requires an accurate and timely assessment of the tumor's response to a particular treatment. A computed tomography (CT) scan can be used to assess the effectiveness of a treatment by evaluating the complete response as the removal of breast lesions based on the Response Evaluation Criteria In Solid Tumors (RECIST) classification. Following the surgical excision of any remaining tumors and lymph nodes, the response to treatment in early-stage breast cancer treated with neoadjuvant chemotherapy (NAC) can be assessed pathologically. Under such circumstances, the pathological complete response (pCR), as defined by, among other classifications, Chevalier's or Sataloff's [9], is characterized by the lack of residual tumor in the breast and related axillary lymph nodes. By assigning a score to the residual illness, the Residual Cancer Burden (RCB) index more accurately assesses the response to neoadjuvant chemotherapy, making it possible to differentiate between treatment resistance and partial response [10]. pCR can be utilized to both direct the adjuvant therapy and validate a biomarker. Only individuals with an illness that persisted after NAC could receive further chemotherapy in an adjuvant setting in the KATHERINE and CREATE-X prospective investigations [11]. In patients who achieved a pCR, response to NAC was also used to prevent needless further chemotherapy (COMPASS-HER2 and DESCRESCENDO clinical studies NCT04266249, NCT04675827).

The most helpful biomarkers for breast cancer to date for prognosis and prediction of response to hormonal therapy and targeted therapy, respectively, are ER and HER2. Targeted therapy can be driven by genomic abnormalities, such as BRCA1/2 mutations or PI3KCA mutations, which have also been verified as actionable biomarkers [12]. However, new prognostic biomarkers are still required, especially for ER- and HER2-positive tumors that acquire resistance mechanisms, as well as triple-negative breast cancers. Few were incorporated into clinical practice despite being validated in preclinical investigations. In order to identify predictive biomarkers (molecular or imaging biomarkers) with better accuracy, novel methodologies have been taken into consideration (Figure 3).

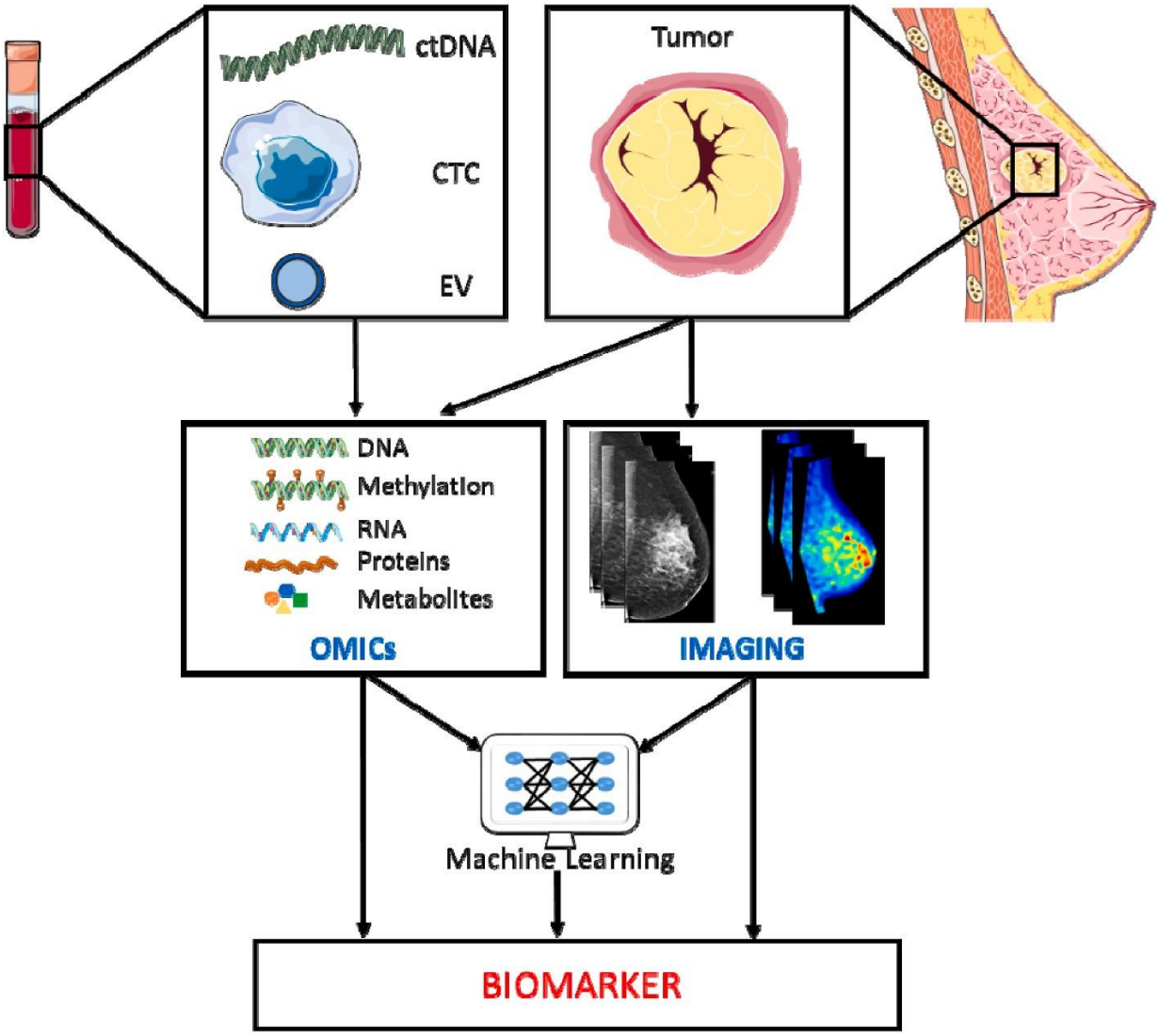

Fig. 3. Technical methods for identifying biomarkers. Molecular (OMICs) and morphologic (IMAGING) data are obtained from solid biopsies (tumor) and liquid biopsies (blood that contains ctDNA, CTC, and EV). Relevant biomarkers are found through analysis of these data, whether or not integrated techniques including machine learning algorithms are used [3]. CTC: Circulating Tumor Cell; EV: Extracellular Vesicle; ctDNA: Circulating Tumor DNA.

4. Blood-Based Biomarkers

Circulating tumor cells (CTCs) and circulating tumor DNA (ctDNA), which can both passively enter the circulation, are examples of blood-based biomarkers. It has been demonstrated that CTC counts or CTC molecular characterization using qPCR can predict therapeutic resistance. The characterization of metastatic disease that is linked to patient outcome is further aided by single cell RNA sequencing, which could enhance patient stratification and therapy approaches [3,13]. It's interesting to note that Copy Number Alteration (CNA) analysis can reveal chemo-resistant clones that may surface during therapy. The response to HER2-targeting therapy is predicted by both CNAs and HER2 copy numbers. A straightforward blood test may find clinical use in the future as ctDNA measurement is more straightforward than CTC counts and serves as a reliable indicator of treatment response in metastatic breast tumors [3]. Murtaza et al. provided evidence that ctDNA sequencing can be used to detect mutations linked to treatment resistance development in invasive tumors. Genomic analysis of ctDNA from 31 patients with breast cancer revealed information about gene changes that underlie treatment resistance in a retrospective research [14]. In a phase 2 clinical trial, ctDNA was also assessed as a predictive biomarker of response to immunotherapy, and a change in ctDNA levels from the baseline was predictive of therapeutic efficacy. ctDNA was tracked in the cTRAK-TN experiment to identify tumor recurrence early and direct additional treatment [3]. However, the therapeutic usefulness of ctDNA for early tumor recurrence or metastasis diagnosis was limited in this investigation since detectable levels of ctDNA were linked to detectable illness. The potential of ctDNA for early molecular relapse identification in patients before any clinically evident illness on imaging is being evaluated by ongoing trials like TRAK-ER (NCT04985266) [15]. The clinical usefulness of ctDNA measures on improving patient outcomes will be determined based on the findings.

The prognostication of early-stage breast cancer using CTC enumeration has been assessed. In a prospective investigation involving blood samples from 487 patients with breast cancer, Tkaczuk and colleagues found CTCs in 56% of the patients, with 83% of those patients having stage IV illness [16]. More than ten CTCs per sample were linked to a lower chance of survival. Other large multicenter trials have demonstrated that the presence of CTCs five years after the end of chemotherapy was predictive of poor response to treatment (RFS) and that increased CTCs at primary diagnosis are related to shorter DFS and OS. Remarkably, a

retrospective study by Goodman et al. proposed that the effectiveness of adjuvant radiation therapy in individuals with early-stage breast cancer is predicted by CTC identification following surgery. Although further prospective interventional trials are required to confirm these findings, they demonstrate the potential of CTC to identify latent micro-metastases, also known as minimal residual disease (MRD), in cases of breast cancer [5].

5. Ki67 IHC

IHC may measure the nuclear protein marker of cell proliferation, Ki67 (Figure 4), and multiple studies indicate a negative correlation between higher expression and prognosis.

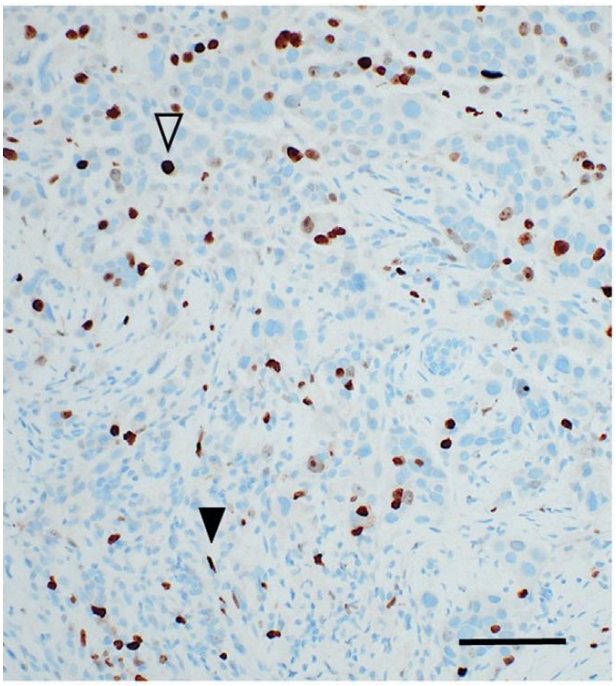

Fig. 4. An invasive breast cancer's associated H&E picture [6]. It is important to distinguish between tumor cells (open arrowhead) that are positive for Ki67 and stromal cells (filled arrowhead) and inflammatory cells.

However, because of substantial interobserver variability, disagreements over scoring, and differences in pre-analytic parameters between laboratories, the systematic use of Ki67 IHC in breast cancer research is contentious [17]. It's important to score Ki67 carefully, assessing just tumor cells and ignoring stroma or inflammatory cells in the surrounding area. Updated guidelines for using Ki67 have been released by the International Ki67 in Breast Cancer Working Group, and they incorporate a uniform visual grading system. Eventually, they came to the conclusion that the

very limited clinical utility of Ki67 IHC is to avoid the necessity for MGA testing in women whose early-stage tumors are ER-positive and have Ki67 levels of ≤5% or ≥30% (since there is no observer concordance at intermediate levels). However, the FDA approved the CDK4/6 inhibitor abemaciclib (Verzenio) in 2021 as an adjuvant treatment for patients with a Ki67score of ≥20% who are ER-positive, HER2-negative, and node-positive and at high risk of recurrence when combined with endocrine therapy. This was predicated on the monarchE trial's preliminary findings [18]. Patients with ≥4 positive axillary lymph nodes, or 1-3 nodes if the tumor is ≥5 cm or grade 3, and Ki67 ≥20% are the only ones eligible for FDA approval. On the basis of the study results, American Society of Clinical Oncology (ASCO), however, suggests abemaciclib plus endocrine therapy for a wider group of patients with ≥4 positive nodes or 1-3 positive nodes, and either grade 3 illness, tumor size ≥5cm or Ki67index ≥20%. It's interesting to note that the advantage of abemaciclib was seen in a follow-up study of the monarchE trial, irrespective of Ki67 index. It is unclear if recommendations and indications for the treatment will alter when more follow-up data becomes available. The Ki67 IHC MIB-1 pharmDx, which is conducted on the Agilent Dako Omnis platform and was utilized in the central laboratory of the monarchE trial, is the FDA companion diagnostic assay for abemaciclib. In the event that a clinical laboratory is not currently using this platform, they might think about cross-validating a test created in-house or submitting this assay to a commercial laboratory [6,19].

6. Circulating Cell-Free DNA (ccfDNA)

The phrase "liquid biopsy" describes both circulating ccfDNA, including ctDNA, and CTCs. To be more precise, circulating DNA is released into the bloodstream by both tumor and normal cells. It is believed to be caused by tumor necrosis, apoptosis, lysis of circulating cancer cells, and/or the release of DNA from rapidly dividing cells [20]. Note that circulating DNA from tumor cells is referred to as ctDNA, whereas free DNA of any origin circulating in the bloodstream is referred to as ccfDNA. Although ccfDNA is found in healthy controls, cancer patients—including those with breast cancer—have more of it than healthy ones do. There are known correlations between ccfDNA levels and nodal involvement, tumor size, and cancer stage. Panagopoulou et al. showed that ccfDNA quantification is a prognostic sign for progression-free survival (PFS) in metastatic illness specifically, and their machine-learning-driven study pinpointed ccfDNA quantification as a powerful predictive marker for response to first-line

treatment [21]. These findings are consistent with a meta-analysis conducted by Tan et al., which discovered that ccfDNA quantification had prognostic value in both early breast cancer and metastatic disease. Nevertheless, the authors acknowledged that there was significant heterogeneity between studies because of variations in analysis type, sampling time, and sampling method, and that their statistical analysis may have overestimated ccfDNA's prognostic role [5].

7. Breast Cancer Stem Cells (BCSCs)

Breast cancer stem cells (BCSCs) induce tumors and promote self-renewal, which both contribute to the disease's aggressive progression and recurrence. Breast cancer tumor growth, metastasis, and resistance to conventional therapy are mostly caused by breast cancer stem cells, which possess the characteristics of tumor-initiating cells (TICs) [22]. Aggressive, varied, and treatment-resistant cancers are frequently associated with these cells [23].

Although recent developments in targeting BCSCs have demonstrated promise in raising remission rates, tumor heterogeneity continues to pose a challenge for targeted therapy. Cancer stem cells (CSCs) are tumor cells that have the capacity to proliferate. DNA damaging agents such as chemotherapy or radiation can induce senescent phenotype in both normal and cancer cells. This therapy-induced senescence (TIS) affects a tiny fraction of tumor cells, and it is an extremely heterogeneous and dynamic process. Although senescent cancer cells could be eradicated by the immune system, they may regain some stemness characteristics leading to tumor growth or cancer recurrence. Treatment-resistant CSCs have the potential to cause tumor relapses [24]. Studies demonstrating that xeno-transplanted cell subpopulations enriched for CSCs can produce tumors from a tiny fraction of unselected cells indicate that CSCs are present in breast cancer [23].

Since stem cell-derived markers have the potential to greatly enhance disease specificity and allow for customized treatment regimens and stem cell-targeted therapy, BCSCs hold great promise for optimizing the treatment of breast cancer by acting as biomarkers for existing therapeutic approaches.

Since single-cell transcriptomics and single-cell genetic lineage tracing became available, the field of inquiry on BCSC origins has grown. The dedifferentiation of adult mammary cells into stem cells through the epithelial-to-mesenchymal transition, the production of BCSCs from normal

stem cells, and the progenitor cells' mutation-induced pluripotency into cancer stem cells (Figure 5) are the main theories regarding the development of BCSCs [25].

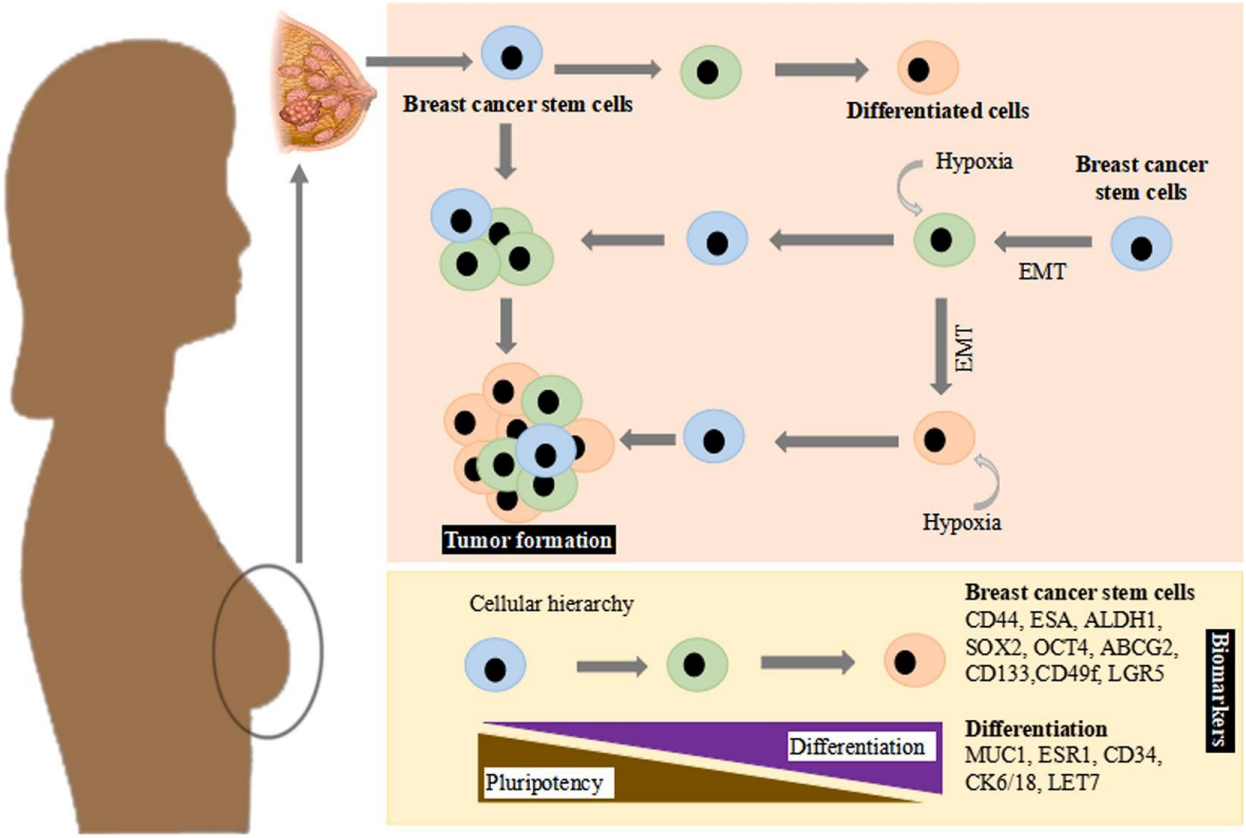

Fig. 5. The cellular ladder from differentiated cells to BCSCs [23].

Since their identification in 2003, BCSCs have been detected using a variety of biomarkers discovered in tumor samples, animal models, and cell lines [25]. These biomarkers show variation in several subgroups among BC subtypes, which is correlated with treatment responses and clinical outcomes. Cell surface markers CD44 and CD24 were originally used by Al-Hajj and associates (2003) to identify tumor-initiating CSCs in BC [23]. Other markers such ABCG2, CD133, CD49f, LGR5, SSEA-3, CD70, and PROCR have also been utilized in recent research to describe BCSCs. The most popular biomarkers for determining BCSC features are CD44, CD24, and ALDH1 [25,26]. Studies have demonstrated that BCSCs exhibiting the CD44+/CD24−/low phenotype and ALDH1+ are responsible for drug resistance, metastasis, tumor start, and progression [27].

For the detection of BCSCs, other less often utilized biomarkers include CD90, PCOR, CD61, CD133, and CD49f. In invasive BC, overexpression of CD133 is associated with a worse

prognosis. When cancer cells are subjected to chemicals that damage DNA, such as radiation and chemotherapy, a process known as senescence takes place. It can promote the growth of cancer stem cells and activate tumor suppressor pathways such as p53. p53 protein expression, nuclear p53 concentration, beta-galactosidase activity, and HP1 gamma are examples of traditional biomarkers of cellular senescence [28]. Different signaling mechanisms govern the stemness, self-renewal, metastasis, and treatment resistance of BCSCs (Figure 6).

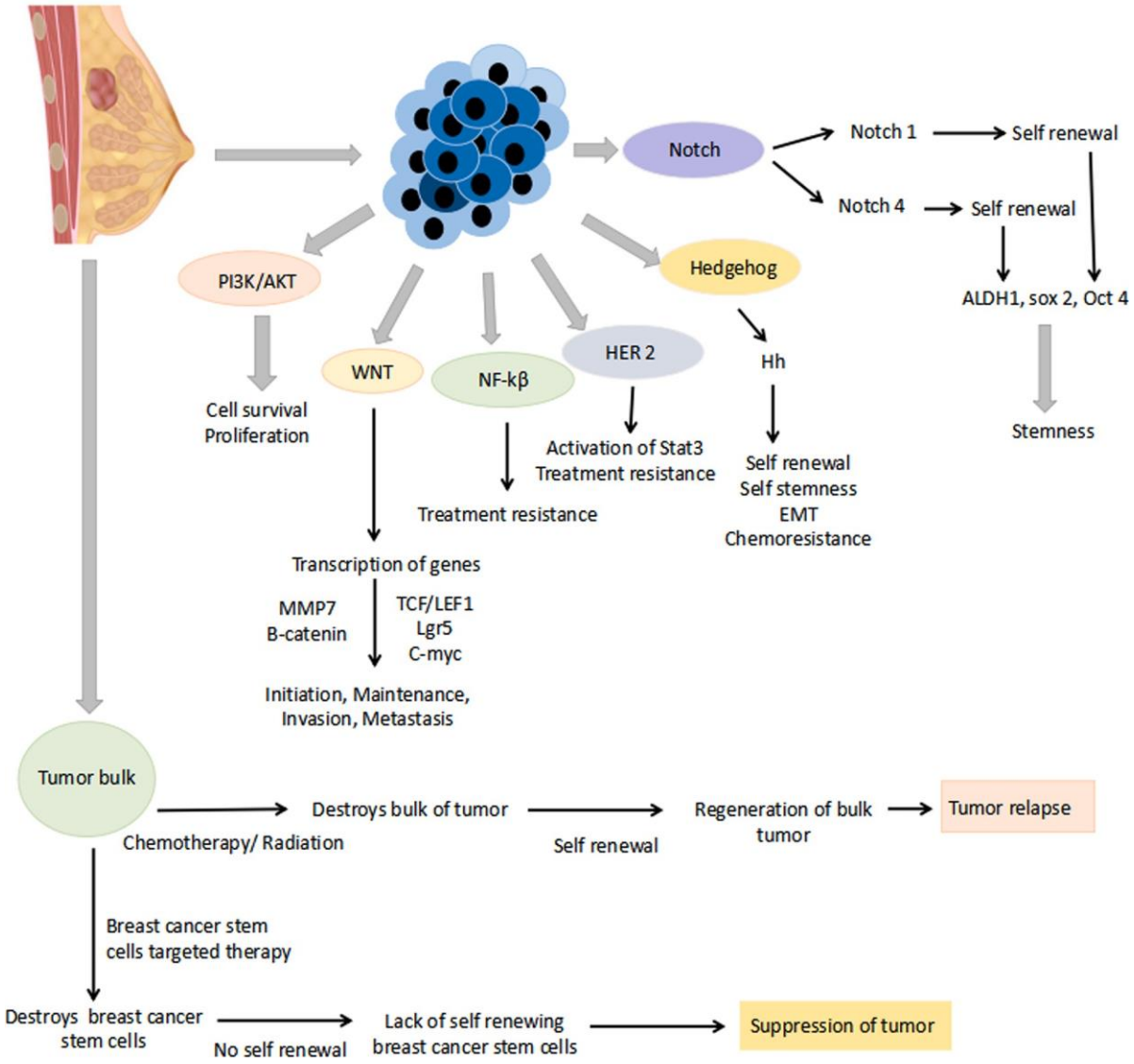

Fig. 6. Various substances and signaling pathways in the control of BCSCS to the phenomena of tumor suppression and recurrence [23].

Genetic abnormalities or other deregulations of these pathways can lead to the transformation of normal stem cells into BCSCs, which subsequently multiply uncontrollably to create cancers. In addition to these signaling pathways, non-coding RNAs (ncRNAs), mainly microRNAs (miRNAs) and long non-coding RNAs (lincRNAs), are important regulators in BCSCs [29].

8. BCSCS' Function as Biomarkers for Precision Medicine in Breast Cancer

Precision medicine refers to a method of treating illnesses that takes individual variations in genetics, environment, and lifestyle into account resulting in more beneficial outcomes [30], Figure 7. Increasing precision in precision medicine has been the aim of recent developments in omics technologies. The discovery of relevant biomarkers to predict the efficacy of customized therapy in a specific patient population forms the basis of the precision-medicine approach. In breast cancer, certain druggable mutations have been discovered. New therapeutic modalities are being investigated in clinical trials for patients with metastatic cancer, specifically for BCSCs. Using cell surface marker molecules or dye-efflux sub-population isolation approaches, CSCs have been isolated from a variety of cancer types. Research indicates that tumors expressing higher amounts of CD44 and CD133 are more likely to develop early liver metastases, which further strengthens their significance as predictive indicators of poor prognosis and potential therapeutic targets. Early in the course of treatment, predictive biomarkers can reliably evaluate the efficacy of anticancer treatment options, enabling the most appropriate therapeutic intervention to be chosen as well as the necessary modifications to treatment regimens [31]. To better tailor anticancer treatment, more data on "interpersonal disease variations" can be obtained by examining patient groups with low treatment-outcome prediction rates. The discovery of distinct biochemical and metabolic characteristics of CSCs, as well as the identification of stem cell markers in vivo and in vitro, have revealed their important role in the development, metastasis, chemoresistance, and tumor recurrence of cancer [32]. Studies have demonstrated the variability of human bone marrow cells' reactions to various anticancer drugs, and there is a correlation between the anticancer drugs' chemotherapeutic effects and the molecular markers expressed by BCSCs. The potential application of CSCs and their markers in pre- and post-chemotherapy initiation is suggested by their possible use in customizing treatment regimens for breast neoplasia [23]. When it comes to aspects of anti-cancer treatment such as tumorigenicity, metastasis,

chemo/radioresistance, and multidrug resistance, CSCs are essential. Molecular indicators of identification and differentiation as well as options for targeted treatment of CSCs have been the focus of research. CSC indicators can be useful tools for treating patients according to their needs and may be essential for their identification or targeting. The expression levels of multiple molecular markers of BCSCs are connected with the variability in the chemotherapeutic/cytotoxic action of anticancer drugs in human BC cells, indicating the potential utility of these markers as biomarkers in the fight against BC [23].

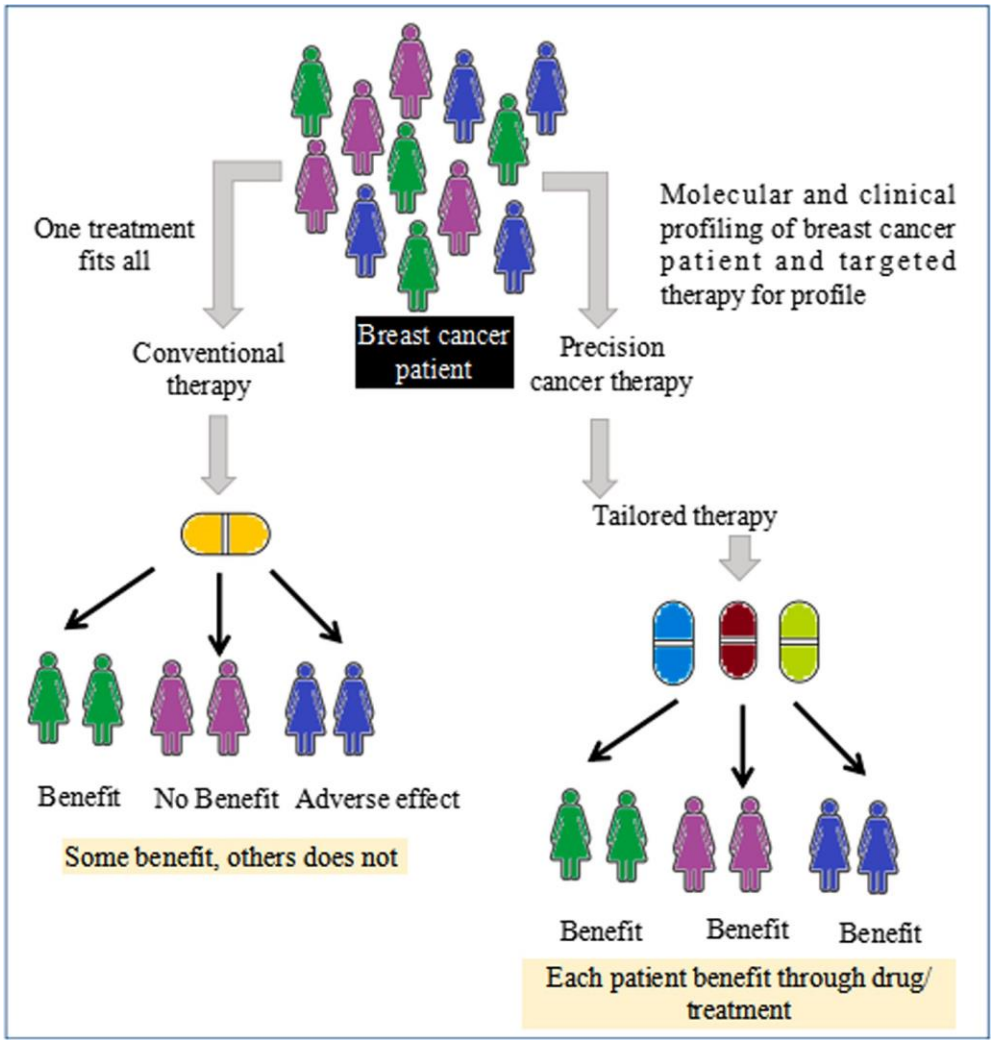

Fig. 7. What part do BCSCs play in precision medicine for treating BC? [23]

9. Conclusion: Future Perspectives

Breast cancer remains a complex and prevalent disease that requires ongoing advancements in biomarkers for optimized management. This review article has provided a comprehensive

overview of the progress made in breast cancer biomarkers and their implications in diagnosis, prognosis, and treatment decision-making. By exploring both traditional and emerging biomarkers, as well as their integration into precision medicine approaches, this article highlights the significant strides made in improving breast cancer management. The progress achieved in breast cancer biomarkers has been remarkable and has revolutionized the field of breast cancer management. Traditional biomarkers, such as hormone receptors and HER2, have played a critical role in guiding treatment decisions and predicting patient outcomes. Their assessment has allowed for tailored therapeutic strategies, leading to improved patient outcomes and reduced unnecessary treatments. In addition to traditional biomarkers, emerging biomarkers have shown great promise in advancing breast cancer management. The detection and analysis of circulating tumor cells (CTCs) and liquid biopsies, such as circulating tumor DNA (ctDNA), offer non-invasive and real-time monitoring of tumor dynamics, treatment response, and minimal residual disease. These biomarkers have the potential to improve early detection, guide treatment decisions, and monitor treatment effectiveness, ultimately contributing to optimized patient management. The integration of biomarkers into precision medicine has further enhanced breast cancer management. Personalized treatment options based on biomarker profiles have transformed the therapeutic landscape, allowing for targeted therapies and immunotherapies tailored to individual patients. Biomarker-driven clinical trials have not only accelerated the development of novel therapies but also facilitated the identification of predictive biomarkers for treatment response and resistance, paving the way for more effective and individualized treatment strategies. Despite the significant progress made, challenges remain on the path toward optimized breast cancer management. The translation of biomarker discoveries into routine clinical practice requires addressing issues related to regulatory approval, standardization, and widespread accessibility. Additionally, the economic considerations and ethical implications associated with biomarker testing and treatment decisions need careful consideration. Looking ahead, continued research and collaboration are essential to further advance breast cancer biomarkers. Integration of multi-omics data, including genomics, transcriptomics, proteomics, and epigenomics, holds promise in unravelling the underlying mechanisms of breast cancer and identifying novel therapeutic targets. Advancements in artificial intelligence and machine learning algorithms can aid in the discovery of new biomarkers, improve diagnostic accuracy, and predict treatment response. Furthermore, ongoing efforts to enhance patient education and engagement are crucial to ensuring that the benefits of biomarker-based

management strategies are effectively communicated and implemented. To emphasize, significant progress has been made in breast cancer biomarkers, leading to optimized management strategies. While challenges persist, the integration of traditional and emerging biomarkers into precision medicine approaches has paved the way for personalized treatment options and improved patient outcomes. Continued research, collaboration, and innovation are necessary to overcome the remaining hurdles and achieve the ultimate goal of optimizing breast cancer management. By harnessing the potential of biomarkers, we can work toward a future where breast cancer is detected earlier, treated more effectively, and ultimately, where lives are saved.

- **Conflict of Interest**

The authors declare that they have no competing interests.

References

1. Perou CM, Sørlie T, Eisen MB, van de Rijn M, Jeffrey SS, Rees CA, et al. Molecular portraits of human breast tumours. Nature. 2000 Aug 17;406(6797):747–52.
2. Parker JS, Mullins M, Cheang MCU, Leung S, Voduc D, Vickery T, et al. Supervised Risk Predictor of Breast Cancer Based on Intrinsic Subtypes. J Clin Oncol. 2009 Mar 10;27(8):1160–7.
3. Rodrigues-Ferreira S, Nahmias C. Predictive biomarkers for personalized medicine in breast cancer. Cancer Lett. 2022;545(April):215828.
4. Khan MM, Yalamarty SSK, Rajmalani BA, Filipczak N, Torchilin VP. Recent strategies to overcome breast cancer resistance. Crit Rev Oncol Hematol. 2024 May;197(April):104351.
5. Seale KN, Tkaczuk KHR. Circulating Biomarkers in Breast Cancer. Clin Breast Cancer. 2022;22(3):e319–31.
6. Almajnooni A, Laharwani H, Wu A, Young AL, Lin CY, Sun L. Predictive and prognostic biomarker testing in invasive breast cancer. Diagnostic Histopathol. 2023;29(3):162–72.
7. Wang X, Zhang H, Chen X. Drug resistance and combating drug resistance in cancer. Cancer Drug Resist. 2019 Jun 19;2(2):141–60.
8. Beelen K, Zwart W, Linn SC. Can predictive biomarkers in breast cancer guide adjuvant endocrine therapy? Nat Rev Clin Oncol. 2012 Sep 24;9(9):529–41.
9. Abrial C, Thivat E, Tacca O, Durando X, Mouret-Reynier MA, Gimbergues P, et al.

Measurement of Residual Disease After Neoadjuvant Chemotherapy. J Clin Oncol. 2008 Jun 20;26(18):3094–3094.

10. Symmans WF, Peintinger F, Hatzis C, Rajan R, Kuerer H, Valero V, et al. Measurement of Residual Breast Cancer Burden to Predict Survival After Neoadjuvant Chemotherapy. J Clin Oncol. 2007 Oct 1;25(28):4414–22.
11. Masuda N, Lee SJ, Ohtani S, Im YH, Lee ES, Yokota I, et al. Adjuvant Capecitabine for Breast Cancer after Preoperative Chemotherapy. N Engl J Med. 2017 Jun 1;376(22):2147–59.
12. Crimini E, Repetto M, Aftimos P, Botticelli A, Marchetti P, Curigliano G. Precision medicine in breast cancer: From clinical trials to clinical practice. Cancer Treat Rev. 2021 Jul 1;98:102223.
13. Danenberg E, Bardwell H, Zanotelli VRT, Provenzano E, Chin SF, Rueda OM, et al. Breast tumor microenvironment structures are associated with genomic features and clinical outcome. Nat Genet. 2022 May 18;54(5):660–9.
14. Chen Z, Sun T, Yang Z, Zheng Y, Yu R, Wu X, et al. Monitoring treatment efficacy and resistance in breast cancer patients via circulating tumor DNA genomic profiling. Mol Genet Genomic Med. 2020 Feb 23;8(2):e1079.
15. Garcia-Murillas I, Chopra N, Comino-Méndez I, Beaney M, Tovey H, Cutts RJ, et al. Assessment of Molecular Relapse Detection in Early-Stage Breast Cancer. JAMA Oncol. 2019 Oct 1;5(10):1473.
16. Tkaczuk KHR, Goloubeva O, Tait NS, Feldman F, Tan M, Lum ZP, et al. The significance of circulating epithelial cells in Breast Cancer patients by a novel negative selection method. Breast Cancer Res Treat. 2008 Sep 7;111(2):355–64.
17. Polley MYC, Leung SCY, McShane LM, Gao D, Hugh JC, Mastropasqua MG, et al. An International Ki67 Reproducibility Study. JNCI J Natl Cancer Inst. 2013 Dec 18;105(24):1897–906.
18. Johnston SRD, Harbeck N, Hegg R, Toi M, Martin M, Shao ZM, et al. Abemaciclib Combined With Endocrine Therapy for the Adjuvant Treatment of HR+, HER2−, Node-Positive, High-Risk, Early Breast Cancer (monarchE). J Clin Oncol. 2020 Dec 1;38(34):3987–98.
19. Raab R, Ismaila N, Andre F, Stearns V, Kalinsky K. Biomarkers for Adjuvant Endocrine

and Chemotherapy in Early-Stage Breast Cancer: ASCO Guideline Update Q and A. JCO Oncol Pract. 2022 Sep 21;18(9):646–8.

20. Diehl F, Li M, Dressman D, He Y, Shen D, Szabo S, et al. Detection and quantification of mutations in the plasma of patients with colorectal tumors. Proc Natl Acad Sci. 2005 Nov 8;102(45):16368–73.
21. Panagopoulou M, Karaglani M, Balgkouranidou I, Biziota E, Koukaki T, Karamitrousis E, et al. Circulating cell-free DNA in breast cancer: size profiling, levels, and methylation patterns lead to prognostic and predictive classifiers. Oncogene. 2019 May 14;38(18):3387–401.
22. Walcher L, Kistenmacher AK, Suo H, Kitte R, Dluczek S, Strauß A, et al. Cancer Stem Cells—Origins and Biomarkers: Perspectives for Targeted Personalized Therapies. Front Immunol. 2020 Aug 7;11:1280.
23. Ray SK, Mukherjee S. Breast cancer stem cells as novel biomarkers. Clin Chim Acta. 2024;557(March):117855.
24. Liao C, Xiao Y, Liu L. The Dynamic Process and Its Dual Effects on Tumors of Therapy-Induced Senescence. Cancer Manag Res. 2020 Dec 31;Volume 12:13553–66.
25. Zhang X, Powell K, Li L. Breast Cancer Stem Cells: Biomarkers, Identification and Isolation Methods, Regulating Mechanisms, Cellular Origin, and Beyond. Cancers (Basel). 2020 Dec 14;12(12):3765.
26. Tsintarakis A, Papalouka C, Kontarini C, Zoumpourlis P, Karakostis K, Adamaki M, et al. The Intricate Interplay between Cancer Stem Cells and Oncogenic miRNAs in Breast Cancer Progression and Metastasis. Life. 2023 Jun 9;13(6):1361.
27. He L, Wick N, Germans SK, Peng Y. The Role of Breast Cancer Stem Cells in Chemoresistance and Metastasis in Triple-Negative Breast Cancer. Cancers (Basel). 2021 Dec 9;13(24):6209.
28. Joseph C, Arshad M, Kurozomi S, Althobiti M, Miligy IM, Al-izzi S, et al. Overexpression of the cancer stem cell marker CD133 confers a poor prognosis in invasive breast cancer. Breast Cancer Res Treat. 2019 Apr 15;174(2):387–99.
29. Flores-Huerta N, Silva-Cázares MB, Arriaga-Pizano LA, Prieto-Chávez JL, López-Camarillo C. LncRNAs and microRNAs as Essential Regulators of Stemness in Breast Cancer Stem Cells. Biomolecules. 2021 Mar 3;11(3):380.

30. Johnson KB, Wei W, Weeraratne D, Frisse ME, Misulis K, Rhee K, et al. Precision Medicine, AI, and the Future of Personalized Health Care. Clin Transl Sci. 2021 Jan 12;14(1):86–93.
31. Ray SK, Mukherjee S. LncRNAs as Architects in Cancer Biomarkers with Interface of Epitranscriptomics- Incipient Targets in Cancer Therapy. Curr Cancer Drug Targets. 2021 May 8;21(5):416–27.
32. Ahmed N, Kadife E, Raza A, Short M, Jubinsky PT, Kannourakis G. Ovarian Cancer, Cancer Stem Cells and Current Treatment Strategies: A Potential Role of Magmas in the Current Treatment Methods. Cells. 2020 Mar 14;9(3):719.