

Review article

Drug resistance in breast cancer

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ABSTRACT

Objective: The obstacle of resistance throughout breast cancer treatment leads to failure of therapy and progress in the disease. Current developments in our knowledge of the molecular pathways behind resistance to therapies in breast cancer will be covered in this brief review.

Materials and methods: The presented data in this review is gathered from multiple sources such as clinical observations (information about how patients react to systemic treatments), experimental models (data gathered from in vitro and in vivo drug resistance simulation framework), review of genetic alterations and epigenetic changes that contribute to resistance is done through genomic and epigenetic studies.

The drug resistance mechanisms: target mutations, novel signaling pathways, modifications to the tumor microenvironment, emerging biomarkers, genetic and epigenetic modifications (targeted gene mutations, modifications in drug transporter expression, activation of signaling pathways).

Conclusion: Ongoing investigations are essential for identifying successful approaches to combat drug resistance in breast cancer. The process of refinement of combining medicines, development of novel targeted treatments, and research into mechanisms that limit drug transport are among the areas of focus.

Key words: breast cancer, drug resistance, molecular mechanisms, biomarkers, combination therapies, targeted therapies, drug transport inhibition

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INTRODUCTION

A scenario ought to be pictured where novel research performed using the latest tools and technology provides several useful insights into how cellular resistance as a process works. If strategies could be developed that cause the cells responsible for breast cancer to become sensitized to treatment, then the mysteries behind the phenomenon known as drug resistance could be effectively resolved. This paper engages in an extensive analysis of the various types of molecular intricacies that are involved in the process of drug resistance, and the bargain, language of proteins and genes, and signaling pathways that are responsible for guiding the work of cancer cells will be assessed and well understood. Drug resistance is something that serves as quite an important impediment to the effective treatment of diseases, such as cancer. Despite the many advances and developments that have been made in the field of immunotherapy and targeted therapy, tumors responsible for the occurrence of breast cancer eventually become resistant to the treatment provided for their destruction. This has occurred mainly because the tumor microenvironment has changed, signaling pathways have become activated, and new mutations have developed. This article focuses on gaining an understanding of the unique and intricate nature of the molecular mechanisms that are known to cause resistance to drugs in the tumor cells of patients diagnosed with breast cancer.

CELL LINES AND CULTURE CONDITIONS

Several methods can be used to detect and treat breast cancer. However, the testing process associated with the detection and treatment of breast cancer requires considerable expertise. Innovative technology needs to be used, for instance, to ensure that tumors are correctly identified and that the treatment of tumors which takes place is based on a correct diagnosis. The BT-474, MDA- and MB-231 cell lines were used in this study. Cells were cultured in DMEM/F12 supplemented with 10% FBS, 1% penicillin/streptomycin, and 1% l-glutamine. MDA-MB231 constitutes a cell line that is used quite commonly on the part of those who engage in medical research. When such a hypothesis was tested it was found that the protein data bank deposits become scattered in different parts such as across the cell membrane walls and in the cell cytoplasm. Transmission electron micrographs that were obtained from a treatment program demonstrated that necrotic and apoptotic death were both observed, with each being characterized by a certain range of features. Thus, cell culture methods and techniques are being developed to model cancer structures in 3D.

The heterogeneity in cell lines permits a comprehensive approach to evaluate breast cancer cell lines to obtain a clear

understanding of drug resistance mechanisms. The T-47D cell line has been used in drug resistance research to provide more information regarding hormonal therapies for breast cancer [1]. The application of various cell lines is vital because several subtypes of breast cancer might respond differently to various treatments, and the evaluation of several cell lines helps tailor therapeutic approaches to specific patients [2]. This study reveals that culturing cell lines in a controlled environment is important for reproducibility and testing. DMEM/F12 supplemented with 10% FBS, 1% penicillin/streptomycin, and 1% l-glutamine is an effective culture medium for cell lines and conditions [3]. The development of 3D cell culture strategies has changed this field by providing a more physiologically significant platform for the assessment of drug resistance [4]. Such models resemble the environment of a breast tumor and provide an exact representation of drug response, thereby bridging the gap between traditional monolayer and in vivo cultures as seen in figure 1.

Elucidating the mechanism of drug resistance in breast cancer

To elucidate the mechanism of drug resistance in breast cancer, a complex interplay between various factors is required. These factors include targeted mutations, tumor microenvironment, undiscovered genes, and signaling pathways.

Targeted mutation

Mutations in specific genes can lead to the development of drug resistance in breast cancer. One well-known example is a mutation in the HER2 gene, which can result in resistance to HER2-targeted therapies, such as trastuzumab (herceptin) [5]. Additionally, mutations in genes such as TP53 (p53) and BRCA1/2 can affect DNA repair mechanisms and promote resistance to chemotherapy [6]. Targeted therapies may become less effective as cancer cells evolve to bypass the targeted pathways through mutations.

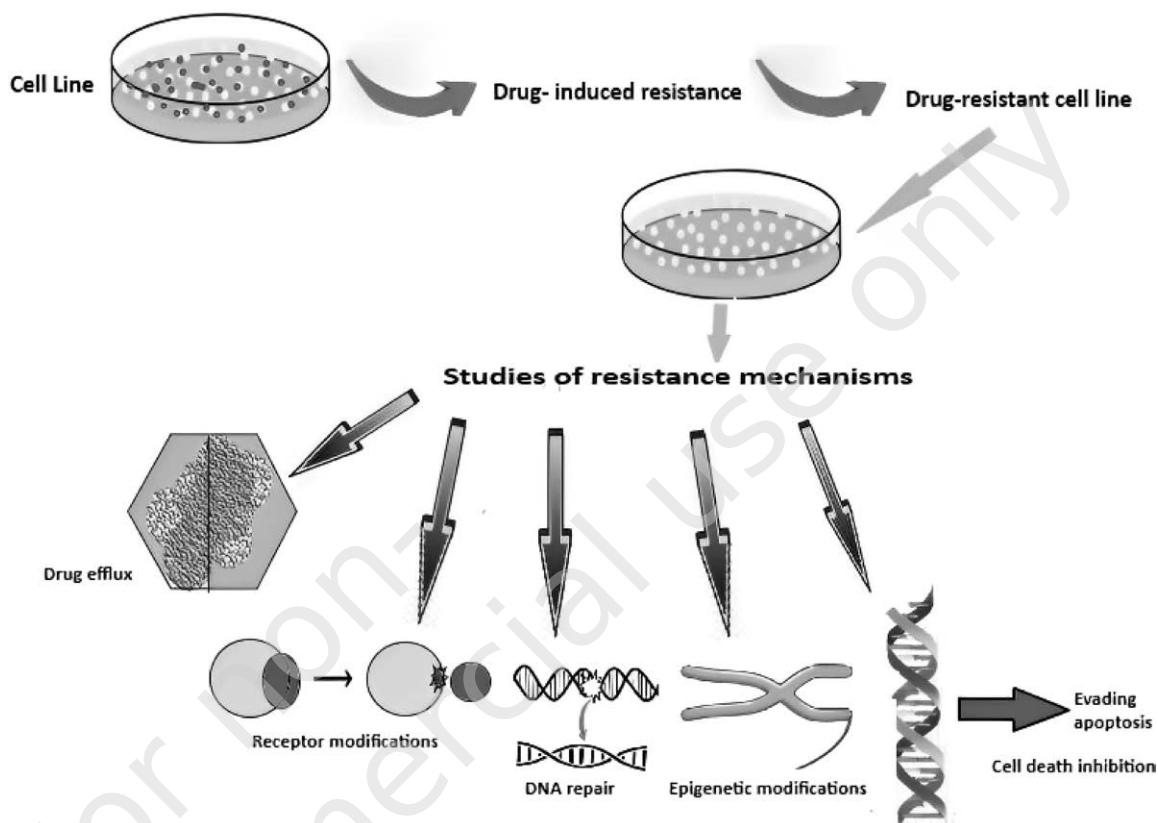
Tumor microenvironment

The tumor microenvironment (TME) plays a crucial role in drug resistance. Factors within the TME, such as immune cells, stromal cells, and extracellular matrix components, can influence cancer cell behavior and response to treatment [7]. TME components secrete factors that promote survival and growth of drug-resistant cancer cells. Hypoxia (low oxygen levels) within the TME can activate the survival pathways that contribute to resistance.

Undiscovered genes

There are likely undiscovered genes that contribute to drug resistance in breast cancer. Developments that have been made in techniques and genomics, such as that of the CRISPR Cas9 method of screening, have made it possible for researchers to

Figure 1. Cell lines to study changes leading to resistance mechanism.



identify genes that play a role in the occurrence of resistance. These genes may be involved in pathways that are not yet well understood or that are not traditionally associated with drug resistance.

Signaling pathways

Various signaling pathways have been implicated in drug resistance. The P13K/AKT/mTOR pathway tends to be dysregulated when breast cancer occurs and is known to cause resistance to drugs, mainly because cell proliferation and survival are promoted. The MAPK/ERK pathway is another example of how a pathway is capable of affecting drug sensitivity. Targeted therapies can also restrict these pathways. However, it must also be noted that resistance can develop in cancer cells through activation of complementary or alternative pathways.

PROMISING DRUG DELIVERY SYSTEM THAT CAN ENHANCE THE SENSITIVITY OF ANTI-BREAST CANCER AGENTS TO VARIOUS TUMORS

Drug delivery systems based on nanoparticles are quite effective and promising, as the sensitivity of breast cancer agents to tumors can be enhanced. These particles were quite small, ranging between 1 and 1000 nm.

Targeted delivery

Thus, it is possible to design nanoparticles that specifically target cancer cells. The tumor microenvironment can also be developed for this purpose. Thus, damage to healthy tissues and cells is something that will be minimized. When nanoparticles are functionalized using targeting ligands, they can selectively bind to cancer cells [7].

Enhanced permeability and retention effect

The enhanced permeability and retention (EPR) effect can be effectively exploited for some nanoparticles. This refers to the tendency of tumors to produce leaky blood vessels, where lymphatic drainage is impaired. This situation makes it possible to increase the concentration of drugs at the tumor site by preferentially accumulating in the tumor tissues [7].

Sustained release

Nanoparticles can encapsulate drugs and release them in a controlled and sustained manner. This can lead to prolonged drug exposure in tumors, enhanced drug efficacy, and reduced side effects. A unique feature of nanoparticles is that drugs can be encapsulated by the same drug and then released in a manner that is sustained and controlled. Prolonged drug exposure is thus something that becomes feasible as a consequence of this,

and the drug administered to control breast cancer is enhanced. These side effects were reduced.

Multidrug loading

Numerous drugs can be administered by nanoparticles, and a combination of different therapies can be administered to different pathways involved in drug resistance.

Overcoming efflux pumps

Nanoparticles can help overcome the drug efflux pumps that cancer cells often use to expel drugs [8]. Encapsulating drugs within nanoparticles makes them less susceptible to efflux, leading to increased intracellular drug accumulation. Drug efflux pumps can be effectively overcome with the help of nanoparticles. These pumps are used by the cancer cells for drug expulsion. If drugs are encapsulated within nanoparticles, their susceptibility to efflux is reduced. Thus, the intracellular accumulation of drugs increased.

Reduced systemic toxicity

By specifically delivering drugs to tumor sites, nanoparticle-based delivery systems can reduce the exposure of healthy tissues to toxic anti-cancer agents, thereby minimizing systemic adverse reactions. As drugs are delivered specifically to the sites of tumors, a drug delivery system based on the use of nanoparticles will help reduce the exposure of healthy tissues to toxic anti-cancer agents. In this process, systematic adverse reactions were considerably reduced.

Personalized medicine

Nanoparticles can be tailored to individual patients based on their specific tumor characteristics and drug responses, enabling a personalized treatment approach. Liposomes and micelles are examples of nanoparticles that can be used for this purpose [8]. These systems have been used to deliver chemotherapeutic drugs, targeted therapies, and nucleic acid-based therapies to breast cancer cells.

Strategies that can improve patient care during bio-chemotherapeutic treatments

During chemotherapy, which combines chemotherapy and immunotherapy, patient care requires a comprehensive and patient-centered approach. The quality of life of patients is strongly affected by the treatment strategy chosen for breast cancer. Fighting spirit, positive reframing, helplessness or hopelessness, and worried obsession are only a few coping strategies documented for cancer. Patients who adopt a positive outlook are inspired to view their condition as a challenge and take steps to

overcome it. Destructive strategies take the form of feelings of helplessness, anxiety, and a propensity to interpret any symptom as a sign of health deterioration, whereas positive redefinition enables patients to find hope and fulfillment in life while maintaining full awareness of the severity of their illness. The latter may increase the negative consequences of mastectomy, particularly the symptoms associated with the breast and arm, and may encourage a submissive attitude toward the condition. Therefore, a system of strategic measures must be included to ensure its effectiveness and efficiency [9, 10].

SMALL MOLECULE COMPOUNDS THAT ARE EFFECTIVE AGAINST DRUG-RESISTANT BREAST TUMORS, BIOMARKERS, OF CHEMOTHERAPY RESISTANCE IN BREAST CANCER PATIENTS

Several molecular compounds have shown promise in targeting drug-resistant breast tumors. Additionally, researchers have identified various biomarkers associated with chemotherapy resistance in breast cancer patients. Below is a discussion of these molecular compounds in table 1:

Table 1. Small molecule and compounds.

Lapatinib	Lapatinib is a tyrosine kinase inhibitor that targets both the HER2 and EGFR receptors. It has shown effectiveness against HER2-positive breast tumors that have developed resistance to other HER2-targeted therapies [11].
Palbociclib, ribociclib, abemaciclib	These CDK4/6 inhibitors have been effective in targeting hormone receptor-positive breast cancer cells, including those resistant to endocrine therapy.
Everolimus	Is an mTOR inhibitor that has shown promise in overcoming resistance to hormone therapy in certain breast cancer cases?
T-DM1 (trastuzumab, emtansine)	T-DM1 is an antibody-drug conjugate that combines trastuzumab with a chemotherapeutic drug. It has demonstrated efficacy in HER2-positive breast cancers resistant to trastuzumab [11].
PARP-inhibitors (olaparib, talazoparib)	PARP inhibitors effectively treat breast cancers with BRCA mutations, which are often associated with increased drug resistance.

Biomarkers of chemotherapy resistance

Table 2. Biomarkers of chemotherapy resistance.

P-glycoprotein (P-gp)	Overexpression of P-gp, a drug efflux pump, reduces the intracellular concentration of chemotherapeutic drugs in cancer cells, leading to drug resistance.
BRCA mutations	Breast cancer patients with mutations in BRCA genes can be more resistant to certain types of chemotherapy but may respond well to PARP inhibitors [11].

TP53 mutations	Breast cancer patients with mutations in BRCA genes can be more resistant to certain types of chemotherapy but may respond well to PARP inhibitors [11].
ERCC1 expression	High expression of ERCC1, a DNA repair enzyme, is associated with resistance to platinum-based chemotherapeutic agents.
Ki-67	High levels of the Ki-67 protein, a cell proliferation marker, have been linked to resistance to hormone therapy and chemotherapy.
HER2 expression	In HER2-positive breast cancer, decreased HER2 expression is associated with resistance to HER2-targeted therapies.
MicroRNA expression	Altered expression of specific microRNAs has correlated with chemotherapy resistance and could serve as predictive biomarkers [11].
DNA repair pathway genes	Alterations in DNA repair pathway genes, such as those involved in homologous recombination repair, can impact sensitivity to DNA-damaging chemotherapy agents.

Table 3. Mechanism of drug resistance in breast cancer.

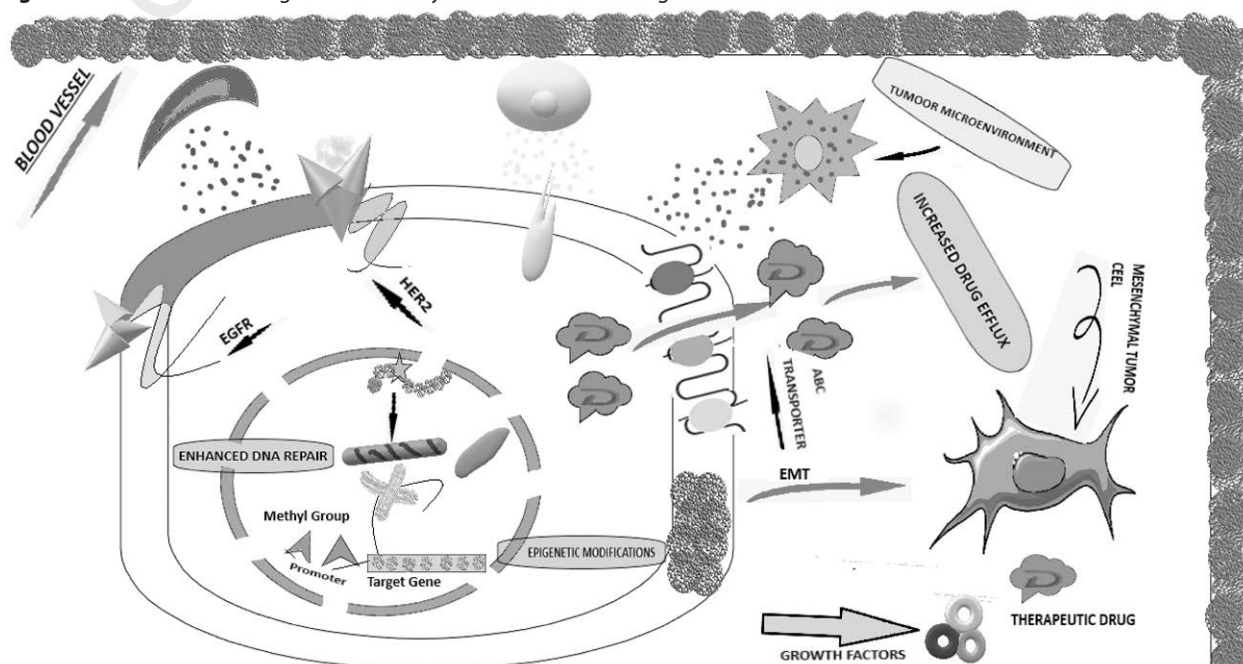
Mechanism	Description
Genetic alterations	Some mutations, like gene-encoding drug targets such as ER, PIK3CA, and HER2 are evident. The primary causes of drug resistance include gene amplification, deletions, and somatic mutations.
Acquired resistance	It involves alterations in drug target genes. Drug resistance can develop as a result of prolonged exposure to treatments, causing genetic changes.
Tumor micro-environment	Interactions with the microenvironment like angiogenesis and tumor-promoting inflammation.
De novo resistance	De novo resistance limits the initial treatment response and may instigate alternative approaches. Some breast cancer cells have drug resistance because of their pre-existing genetic alterations.

Mechanism of drug resistance in breast cancer

Literature reviews revealed numerous molecular mechanisms contributing to drug resistance in breast cancer [12, 13]. Substantial evidence supports the role of genetic alterations, including somatic mutations, gene amplification, and deletions, as the major determinants of resistance to targeted therapies. Mutations in genes encoding drug targets [13]. The encoded drug targets include HER2, ER, and PIK3CA, which are frequently observed in resistant tumors, leading to decreased drug binding or activation of alternate signaling pathways [13]. From this perspective, it is evident that drug resistance in cancer cells is mediated either by acquired or de novo mechanisms as shown in table 3.

Activation of innate mechanisms that guard against harmful foreign chemicals [14], the presence of genetic mutations, and relapse and metastases are common outcomes of chemotherapy owing to multidrug resistance (MDR). Roughly half of all cases of activation of innate mechanisms guard against harmful foreign chemicals [14]; the presence of genetic mutations, the expansion of insensitive subpopulations such as cancer stem cells, and the start of therapy. Acquired resistance, however, may arise through several different mechanisms, including activation of proto-oncogenes, changes in gene expression due to mutations or epigenetic markers, and shifts in the tumor microenvironment. In BC, resistance may occur through several pathways. These include alterations in drug efflux, senescence, DNA repair, tumor heterogeneity, TME, epigenetic changes, and epithelial-to-mesenchymal

Figure 2. Cancer cells showing resistance to cytotoxic anti-cancer drugs.



mal transition (EMT) [14]. The tumor microenvironment, accelerated DNA repair, EMT, epigenetic alterations, and increased drug efflux are all mechanisms of drug resistance in breast cancer as shown in figure 2.

This article highlights the complex interplay between cancer cells and stromal components, such as immune cells, cancer-related fibroblasts, and the therapeutic response matrix, which influence tumor growth, metastasis, and therapeutic response. Immune evasion mechanisms, including up regulation. Immune checkpoint proteins have emerged as important contributors to immunotherapy resistance [15]. Table 4 shows the dysregulated miRNAs related to chemoresistant breast cancer patients based on transcriptome analysis.

Table 4. The dysregulated microRNAs related to chemo-resistant breast cancer patients based on transcriptome scrutiny.

MicroRNA	Expression	Fold Change
Has-miR-195a-5p	upregulated	5.44
Has-miR-4266	upregulated	3.45
Has-miR-200b-3p	upregulated	3.11
Has-miR-214-3p	upregulated	2.99
Has-miR-107	upregulated	2.65
Has-miR-4454	upregulated	2.77
Has-miR-5100	upregulated	2.40
Has-miR-23a-3p	upregulated	2.31
Has-miR-23b-3p	upregulated	2.09
Has-miR-16-5p	upregulated	2.14
Has-miR-4707-5p	downregulated	0.58
Has-miR-3656	downregulated	0.36
Has-miR-1233-1-5p	downregulated	unregulated
Has-miR-3621	downregulated	unregulated
Has-miR-3141	downregulated	unregulated
Has-miR-489	downregulated	unregulated
Has-miR-1227-5p	downregulated	unregulated
Has-miR-1275	downregulated	unregulated
Has-miR-1268b	downregulated	0.41
Has-miR-572	downregulated	0.35
Has-miR-4467	downregulated	0.20
Has-miR-4472	downregulated	0.18

Although RNAs can be associated with chemoresistance, research has noted the necessity of identifying target genes regulated by long noncoding RNAs and the interactions among these genes.

Undiscovered genes and signaling pathways

In addition to the significant advances in understanding drug resistance, this review revealed several gaps in knowledge, especially regarding the undiscovered genes and signaling pathways implicated in the resistance process. Promising preclinical studies have suggested that the involvement of novel genes and reg-

ulatory networks is yet to be fully explored [16]. The findings of these studies underscore the need for comprehensive functional genomic approaches to uncover the hidden molecular drivers of resistance in breast cancer. For instance, a study highlighted that the biological functions of genes in the endocannabinoid signaling pathway, based on protein-protein interactions, were associated with mitochondrial function in major depressive disorder from a genetic and biological function perspective. Table 5 summarizes the seven hub genes mutated in the endocannabinoid pathway in major depressive disorder patients.

Table 5. Seven hub genes mutated in endocannabinoid pathways in patients with major depressive disorder.

Gene	NDUFS4	NDUFV2	NDUFA2	
MD	C0011	D0041	C0025	D0030
Chr	5	5	18	5
Cryptoband	5q11.2	5q11.2	18p1122	Rs79526416
dbSNP	Rs1064793807	Rs886060697	G	T
Ref	GTG	TTTG	C	C
Alt	CTC	-	Splicing	Missense
Clinvar	Non frame shift block substitution	Splicing	NA	Uncertain significance
1KGP	Likely benign	Conflicting interpretations of pathogenicity	NA	0.00079872
ExAC	NA	0.00139776	0.00001653	0.0001
gnomA	NA	0.0032	0.00001219	0.00232
Huabiao project	NA	0.003	0.0001	0.000123

Biomarkers for resistance to systemic therapy

Identifying reliable biomarkers to predict and monitor drug resistance in breast cancer is a critical area of research [18]. This review showed that several potential biomarkers are associated with treatment response and resistance [18]. For instance, circulating tumor DNA (ctDNA) has emerged as a promising non-invasive biomarker that provides real-time information on tumor evolution and acquired mutations [18]. In addition, gene expression signatures and proteomic profiles were investigated to determine their Predictive value for guiding treatment decisions. Table 6 summarizes the characteristics of breast cancer cell lines in suspension culture.

Table 6. Characteristics of the breast cancer cell lines in the suspension culture.

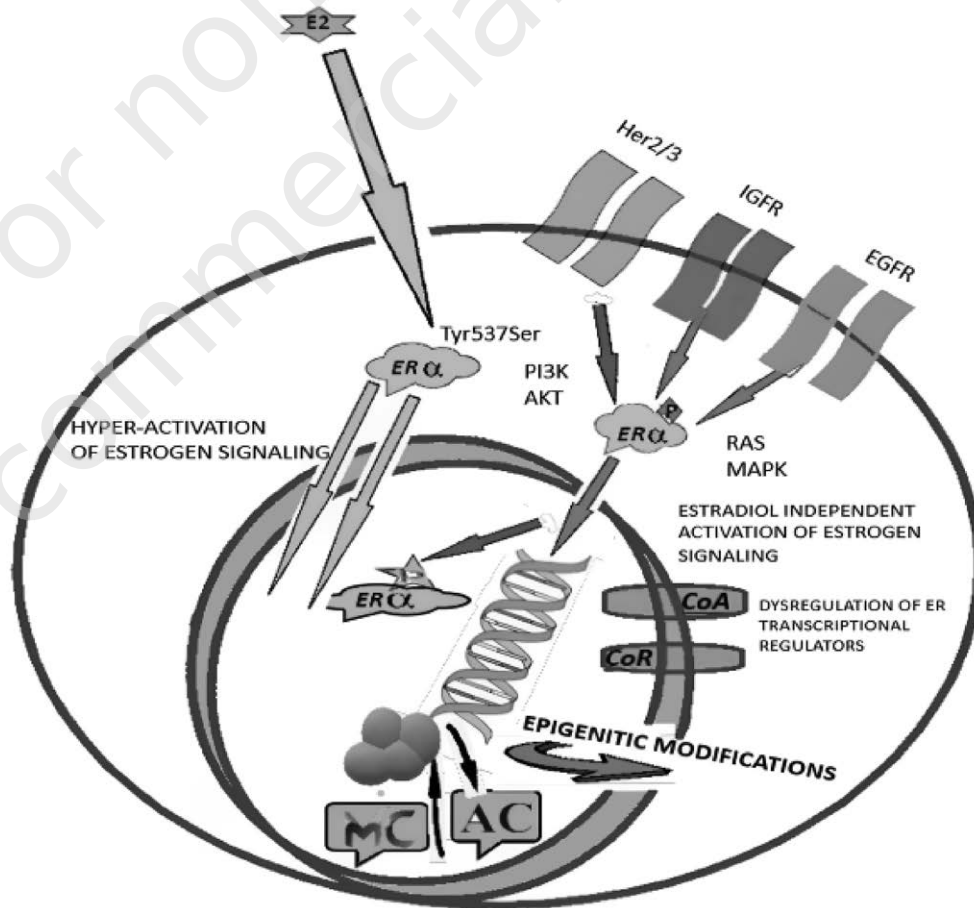
Breast cancer molecular subtypes in suspension culture	Cell line subgroups 5n suspension culture
Luminal A & B	Luminal
Basal-like & claudin-low	Basal A Basal B

Hormonal drug resistance in breast cancer

Hormone receptor-positive (HR+) breast cancer is a common subtype of breast cancer driven by the presence of estrogen and/or progesterone receptors on the surface of cancer cells. These receptors allow cancer cells to respond to hormonal signals that promote growth and survival. Hormone therapy is often used to treat HR+ breast cancers. It blocks the effects of estrogen and suppresses its production, thereby inhibiting the growth of hormone-sensitive cancer cells. However, over time, some HR+ breast cancers have developed resistance to hormone therapy, leading to disease progression and reduced treatment effectiveness. Several mechanisms contribute to the development of hormonal drug resistance in breast cancer. The underlying mechanisms are discussed below in figure 3.

diagnosed worldwide. Research has revealed that ESR1 mutations alter the conformation of ER, producing a constitutively active form of the protein. This study was justified by utilizing a stratified and regulated test for recurrent mutations within the ligand-binding domain of ESR1 in 30% of patients with ER-positive metastatic breast cancer. The identified mutation alterations lower the sensitivity of selective estrogen modulator owing to aromatase inhibitor resistance. There is sufficient research on different types of breast cancers and circulating tumor DNA samples, including other ESR1 fusion genes. The discovery of recurrent ESR1 fusion genes strengthened the notion that resistance to targeted therapies often represents a convergent phenotype. As a result, ESR1 expression was highly affected in cases of resistance to endocrine therapy.

Figure 3. Mechanism of hormonal drug resistance in breast cancer.



ACQUIRED MUTATIONS

Cancer cells can acquire genetic mutations that alter the function of estrogen receptors or related signaling pathways. These mutations make cancer cells less dependent on estrogen for growth, rendering hormone therapy less effective. Research shows that ER-positive breast cancer accounts for 80% of breast cancers

Crosstalk and bypass pathways

Cancer cells can activate alternative signaling pathways that promote growth and survival, bypassing the need for estrogen signaling. For example, some cells may activate growth factor receptors like HER2 (human epidermal growth factor receptor 2) or

other intracellular pathways that can promote cell growth independently of hormonal signaling.

Alterations in estrogen receptor

Changes in the estrogen receptor itself can make it less responsive to hormonal therapy. This might involve mutations, receptor gene amplification, or other modifications that allow the receptor to remain active even in the absence of estrogen.

Micro environmental factors

The tumor microenvironment can influence the response to therapy. Interactions between cancer cells and the surrounding cells, such as immune and stromal cells, can promote resistance by providing survival signals or altering the tumor response to therapy.

Epigenetic changes

Epigenetic modifications, which affect gene expression without altering the underlying DNA sequence, can lead to changes in hormone receptor expression and signaling pathways, contributing to resistance.

Heterogeneity

Tumors are often composed of a mixture of different cancer cell populations. Some cells may be inherently less responsive to hormone therapy or have a higher propensity to develop resistance.

HOW TO OVERCOME HORMONAL DRUG RESISTANCE IN BREAST CANCER?

Therefore, it is essential to understand the molecular pathways that contribute to drug resistance. Evaluation of prognostic indicators as well as clinical and pathological characteristics can inform treatment choices for the management of patients with breast cancer. Although this method is effective, some individuals relapse and/or acquire resistance. Overcoming hormonal drug resistance in breast cancer is challenging; however, several approaches and therapeutic options may be helpful. These include: **Combining treatments:** A combination of different hormonal therapeutic options with a focused mode of treatment can be more effective in the treatment of breast cancer. The use of drugs with different mechanisms of action can help overcome drug resistance. For instance, the combination of an aromatase inhibitor and CDK4/6 inhibitor.

Changing hormonal treatments: Resistance may develop to a particular hormonal therapy, as opposed to the other. Hence, the use of different hormonal therapies may help overcome this resistance.

Targeted therapies: Breast cancer cells may develop resistance through activation of optional pathways. The application of targeted therapies that inhibit these pathways can resolve drug-resistance issues. A good example of this approach is the use of mTOR inhibitors combined with hormonal treatments to treat hormone receptor-positive breast cancer.

Genetic testing: Understanding particular genetic mutations in breast cancer through genetic testing can help in making correct therapeutic decisions. In case of failure, the tumor's genetic profile may be re-evaluated to opt for a new treatment option that aligns with its genetic nature.

Past and current breast cancer clinical trials

BC is present around the milk ducts and lobules. However, it is considered to be in situ; thus, the cancer has not yet manifested and diverged into the rest of the breast. Table 7 summarizes previous and current BC clinical trials concerning HER2, PARP, EGFR, AhR, iNOS, and Wnt.

Table 7. Past and current BC clinical trials concerning HER2, PARP, EGFR, AhR, iNOS, and Wnt.

Intervention/Therapy	Target cancer subtype	Clinical I trial phase	Type	Status	Trial ID reference
KU 0059436 (olaparib), a PARP inhibitor	BRCAL –or BRCA-2-positive advanced BC	phase II	treatment	active	NCT00494234
Preoperative combination of letozole, everolimus, and TRC105e-verolimus and TRC105	Postmenopausal hormone-receptor positive ad Her2BC positive and Her2 BC	phase I	treatment	active	NCT02520063
CDK4/6-inhibitor or	Advanced BC	phase II	treatment	recruiting	NCT03227328
Chemotherapy, in combination with endocrine therapy LGK974 in patients with malignancies dependent on Wnt ligands	TNBC	phase I	treatment	recruiting	NCT01351103

In vitro and in vivo models for studying drug resistance

Preclinical studies have used various in vitro and in vivo models to better understand the complexities of drug resistance in breast cancer. Cell line-based models, patient-derived xenografts (PDX), and genetically modified mouse models (GEMMs) were frequently used to replicate tumor heterogeneity and medica-

tion responses [19]. Insights into the relationship between tumor and stroma and treatment sensitivity were gained using organoid cultures and 3D tumor models. The assessment also admits that none of the models adequately capture the complexity of medication resistance seen in clinical settings.

Emerging strategies for overcoming drug resistance

Innovative strategies for sensitizing breast cancer cells to therapies aimed at overcoming drug resistance have been explored. Combination therapies targeting multiple signaling pathways, exploiting synthetic lethality, and harnessing the potential of the immune system are among the most promising approaches [20]. Targeting resistance-conferring mutations using gene-editing technologies and small-molecule inhibitors has shown encouraging preclinical results, which has motivated further investigation.

Biomarkers for resistance prediction

This review highlights the potential of various biomarkers, including ctDNA, gene expression signatures, and proteomic profiles, for predicting and monitoring drug resistance in breast cancer. These biomarkers hold promise for guiding treatment decisions and facilitating early intervention. Nevertheless, the validation and standardization of these biomarkers in large clinical cohorts remain crucial for their successful translation into clinical practice.

In vitro and in vivo models

The analysis of in vitro and in vivo models emphasizes the importance of using diverse and relevant systems to study drug resistance. Combining different models to complement each other's strengths and weaknesses may provide a more comprehensive understanding of the resistance mechanisms, as shown in table 8. Although these models offer valuable insights, they inherently lack the complexity of the human tumor microenvironment, necessitating caution when translating preclinical findings into clinical settings.

Table 8. In vitro and In vivo models.

In vitro	In vivo
Isolated cellular components are used.	Uses whole living organism
Consumes less time	Consumes more time
Less precise	More precise
Performed under controlled lab conditions	Performed under physiological conditions
Cell structure experiment is in Petri dishes and test tubes	Drug testing experiments are performed through model organisms like mice

Overcoming drug resistance

Exploration of emerging strategies to overcome drug resistance has revealed the potential of combination therapies and immune-based approaches. Rational design of combination regimens targeting multiple pathways and vulnerabilities may effectively circumvent resistance. The promise of gene-editing technologies for targeting resistance-conferring mutations warrants further exploration, although safety and off-target effects remain important considerations.

Implications and limitations

The findings of this study have significant implications in breast cancer treatment and precision medicine. Understanding the diverse molecular drivers of drug resistance can aid in tailoring therapeutic approaches for individual patients, potentially leading to improved outcomes. Moreover, identification of novel biomarkers and therapeutic targets opens new avenues for personalized treatment strategies. However, this study had several limitations. The reliance on published data limits the scope of our analysis to the available literature and the potential for publication bias cannot be disregarded. Additionally, the rapid pace of research in this field may result in outdated findings. Despite our efforts to include the most relevant studies, we have omitted some valuable contributions.

Future directions

Several directions for future research have emerged, based on the identified knowledge gaps and limitations. Large-scale prospective studies are essential to validate the predictive value of these biomarkers in clinical practice.

CONCLUSION

This review comprehensively analyzes the molecular mechanisms underlying drug resistance in breast cancer. These findings highlight the complexity and heterogeneity of resistance mechanisms involving genetic alterations, epigenetic changes, and the tumor microenvironment. The identification of key target mutations, undiscovered genes, and signaling pathways will provide a basis for future research in this area. This review also emphasizes the potential of biomarkers, such as ctDNA and gene expression signatures, to predict and monitor drug resistance. These biomarkers hold promise for guiding treatment decisions and improving patient outcomes. However, further validation and standardization in large clinical cohorts are necessary for its successful implementation in clinical practice. Moreover, our exploration of in vitro and in vivo models has revealed the importance of using diverse and relevant systems to

study drug resistance. Combining different models to complement each other's strengths and weaknesses may enhance our understanding of the resistance mechanisms. Nevertheless, the limitations of preclinical models in fully recapitulating the complexity of drug resistance in patients warrant careful interpretation of the results and cautious clinical translation. The findings of this study have shown that resistance is complex and that the heterogeneous nature of the mechanisms that cause resistance, especially those that involve genetic alterations, tumor microenvironment, and epigenetic transformations, must be taken into consideration.

To understand the mechanisms of drug resistance undiscovered genes, signaling pathways, and key target mutations show that

resistance is a complicated subject and that there is more research that needs to be conducted on the same for a better understanding to be obtained of it. The role that biomarkers such as gene expression signatures and EDNA can play in monitoring and predicting drug resistance is also something that was established in this study. Different types of models are at work insofar as drug resistance is concerned and need to be examined closely to gain a developed notion of how this works. Strategies that can be used to hinder or overcome the type of resistance shown by drug treatments need to be closely evaluated to ensure beneficial outcomes.

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References

1. Amaral MVS, De Sousa Portilho AJ, Da Silva EL et al. Establishment of Drug-resistant Cell Lines as a Model in Experimental Oncology: A Review. *Anticancer Res.* 2019; 39(12): 6443-55.
2. Wu Q, Ba-Alawi W, Deblois G et al. GLUT1 inhibition blocks growth of RB1-positive triple negative breast cancer. *Nat Commun.* 2020; 11(1): 4205.
3. Pivnicka-Worms D. Elucidating mechanisms of farnesyltransferase inhibitor action and resistance in breast cancer by bioluminescence imaging. (2010). <https://doi.org/10.21236/ada540951>.
4. Thompson D. Testing if the SonoTran platform can enhance drug delivery in metastatic colorectal cancer. (2021). <https://doi.org/10.1186/isrctn17598292>.
5. Mesker W. Chemotherapy or Not? Practice-Changing Approach for the Accurate Selection of Patients for Chemotherapy Treatment after Colon Cancer Diagnosis. The Tumour-Stroma-Ratio (TSR) an Addition to the TNM Classification. (2017). <https://doi.org/10.26226/morressier.596dfd-59d462b8029238760d>
6. Luque-Bolivar A, Pérez-Mora E, Villegas VE et al. Resistance and overcoming resistance in breast cancer. *Breast Cancer: Targets Ther.* 2020; 12: 211-29.
7. Pradubayat N, Laoharuangchaiyot J. Molecular mechanisms of drug resistance in breast cancer and potential strategies for overcoming resistance. *Oncología (Ecuador).* 2023; 33: 1-17.
8. Bao X, Zheng W, Sugi NH et al. Small molecule schweinfurthins selectively inhibit cancer cell proliferation and mTOR/AKT signaling by interfering with trans-golgi-network trafficking. *Cancer Biol Ther.* 2015; 16(4): 589-601. <http://doi.org/10.1080/15384047.2015.1019184>.
9. Xu H, Li T, Gong Q et al. Genetic variations in the retrograde endocannabinoid signaling pathway in chinese patients with major depressive disorder. *Front Neurol.* 2023; 14: 1153509.
10. Kinnel B, Singh SK, Oprea-Ilie G, Singh R. Targeted Therapy and Mechanisms of Drug Resistance in Breast Cancer. *Cancers (Basel).* 2023; 15(4): 1320.
11. Pang RW, Poon RT. Clinical Implications of Angiogenesis in Cancers. *Vasc Health Risk Manag.* 2018; 2: 97-108.
12. Liu Y, Zheng C, Huang Y et al. Molecular Mechanisms of Chemo- and Radiotherapy Resistance and the Potential Implications for Cancer Treatment. *MedComm.* 2021; 2: 315-40.
13. Ni Y, Zhou X, Yang J et al. The Role of Tumor-Stroma Interactions in Drug Resistance within the Tumor Microenvironment. *Front Cell Dev Biol.* 2021; 9: 637675.
14. Miyoshi E, Morabito S, Swarup V. Systems Biology Approaches to Unravel the Molecular and Genetic Architecture of Alzheimer's Disease and Related Tauopathies. *Neurobiol Dis.* 2021; 160: 105530.
15. Xu H, Li T, Gong Q et al. Genetic Variations in the Retrograde Endocannabinoid Signaling Pathway in Chinese Patients with Major Depressive Disorder. *Front Neurol.* 2023; 14: 1153509.
16. Haber DA, Velculescu VE. Blood-Based Analyses of Cancer: Circulating Tumor Cells and Circulating Tumor DNA. *Cancer Discovery.* 2014; 4: 650-61.
17. Fiori ME, Di Franco S, Villanova L et al. Cancer-Associated Fibroblasts as Abettors of Tumor Progression at the Crossroads of EMT and Therapy Resistance. *Mol Cancer.* 2019; 18: 1-16.
18. Zhang N, Jiang J, Tang S et al. Predictive Value of Neutrophil-Lymphocyte Ratio and Platelet-Lymphocyte Ratio in Non-Small Cell Lung Cancer Patients Treated with Immune Checkpoint Inhibitors: A Meta-Analysis. *Int Immunopharmacol.* 2020; 85: 106677.
19. Li H, Yang Y, Hong W et al. Applications of Genome Editing Technology in the Targeted Therapy of Human Diseases: Mechanisms, Advances and Prospects. *Signal Transduct. Target Ther.* 2020; 5(1): 1.

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NM and RJ contributed to the design and implementation of the research; NM, RJ, RS, GB, and PT analyzed the results and wrote the manuscript. NM conceived and supervised the project.

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The authors had full access to the data and take full responsibility for its integrity. All authors have read and agreed with the content of the manuscript as written. The paper complies with the Helsinki Declaration, EU Directives and harmonized requirements for biomedical journals.