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Review

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Advanced breast cancer metastasized in the brain: treatment standards and innovations

Elizabeth Klaas, Eric Sung, Esaan Azizi, Melanie Martinez, Arnav Barpujari, Jeffery Roberts, Brandon Lucke-Wold

Department of Neurosurgery, University of Florida, Gainesville, FL 32610, USA.

Correspondence to: Dr. Brandon Lucke-Wold, Department of Neurosurgery, University of Florida, 1505 SW Archer Rd. Gainesville, FL 32608, USA. E-mail: Brandon.Lucke-Wold@neurosurgery.ufl.edu

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Abstract

Breast cancer continues to be a difficult disease to treat due to high rates of metastasis. Metastasis to the brain presents a unique and often overlooked challenge. In this focused review, we discuss the epidemiology of breast cancer and which types frequently metastasize to the brain. Novel treatment approaches are highlighted with supporting scientific evidence. The role of the blood-brain barrier and how it may become altered with metastasis is addressed. We then highlight new innovations for Her2-positive and triple-negative breast cancer. Finally, recent directions for luminal breast cancer are discussed. This review serves to enhance understanding of pathophysiology, spark continued innovation, and provide a user-friendly resource through tables and easy-to-process figures.

Keywords: Breast cancer, innovation, blood-brain barrier, luminal metastasis, new directions

INTRODUCTION

Over the last 25 years, the incidence and mortality of breast cancer has been tremendously increased worldwide[1]. Breast cancer is the most diagnosed cancer in females, with approximately 2.3 million cases and over 680,000 deaths recorded in 185 countries, and is the second leading cause of cancer-related death

after lung cancer[2]. Because of early diagnosis and advances in therapy, the survival rate of patients with breast cancer has greatly increased, with 5-year relative survival up to 90%[3]. However, the rapidly increasing medical costs of tests, surgeries, and top-of-the-line treatments can be almost as devastating as the disease and may be prohibitory in allowing access to the care that is needed for many patients.

Breast cancer is a complex disease that is difficult to treat, especially for patients who are diagnosed with late-stage or metastasized breast cancer. Up to 30% of women diagnosed with early-stage breast cancer later develop metastases, and about 10%-15% of patients with late-stage cancer have brain metastases[1]. These cases require not only consideration of the primary tumor genetics, but also the microenvironment and histology of the metastasized tumors. Despite advances in research and understanding of the disease, breast cancer treatment is still a multifaceted process that may include chemotherapy, immunotherapy, hormonal therapy, radiation, surgery, or a combination of any or all of them. It is important to first understand the type of cancer a patient has before choosing the best method of treatment.

Breast cancer is largely organized into five categories: Luminal A, Luminal B, luminal B-like, triple-negative or basal-like, and human epidermal growth factor receptor 2 enhanced (HER2-positive). More aggressive forms, like triple-negative and HER2-positive, are more likely to metastasize to other organs, including the brain. HER2-positive cancers are likely to respond better to chemotherapeutic or targeted pharmaceutical treatment due to the expressed HER2 protein that can be targeted. Triple-negative cancers do not express any of the common receptors such as HER2, estrogen receptors, or progesterone receptors. This makes triple-negative cancers considerably more difficult to treat due to no specific receptor targets that can be targeted for therapeutic intervention[1].

Luminal A cancers are typically regarded as less aggressive with a good prognosis and account for 73% of breast cancer cases[2,4]. A characteristic of luminal A cancers is low levels of the Ki-67 protein, indicating a slower proliferation rate of cancerous cells. Luminal B type cancers, which account for around 11% of cases, can be notably more aggressive than luminal A cancers. This may be due to a higher proliferation rate of cancerous cells and increased invasive potential for metastasis[1,4]. Luminal B has been linked to a worse prognosis and early recurrence compared to luminal A, potentially due to higher expression of the Ki-67 protein and increased proliferation of cancer cells[5]. Luminal B-like subtype can have any level of Ki-67 protein expression, with progesterone expression that may be positive or negative, and is HER2-positive[1]. These markers indicate poor prognosis when left untreated or diagnosed at advanced disease stages. However, this subtype has a lower risk of metastasizing outside of the breast tissue, which can provide the patient with a slightly better prognosis as the disease may take longer to invade other parts of the body. Both luminal A and B types are estrogen receptor positive, making a prime target for therapeutics[1]. While luminal cancers may have independent levels of risk, both triple-negative and HER2+ cancer types are considered more aggressive than luminal types.

Brain metastases typically originate from malignant cells that reach the brain after traveling via the vascular system from an aggressive primary tumor in the breast. Like the primary tumors, metastatic breast cancer brain tumors are typically treated systemically with chemotherapy or hormonal therapy and may include surgical removal of the tumor(s) [Table 1]. However, because the brain can present obstacles like the blood-brain barrier, treatment for brain metastases may also include more localized approaches like radiotherapy. Luminal cancers respond very well to hormonal therapies that target the receptors they express, and radiation is also commonly used after surgery. However, this could make tumors that metastasize more difficult to treat because metastases may not express the same receptors as the primary tumor, requiring a different approach for treatment as the disease progresses[1].

Table 1. Current drugs used to treat specific subtypes of breast cancer

Type of breast cancer	Treatment	Treatment class	Mechanism of action	Route of administration	Clinical trial	PMID
LUMINAL	Abemaciclib	CDK4/6 inhibitor	Inhibits proteins that signal cell proliferation	Orally	NCT02831530	34226684
LUMINAL	Alpelisib	PI3K Inhibitor	Inhibits tumor cell growth by blocking PIK3 pathway	Orally	NCT02379247	33802424
TRIPLE-NEGATIVE	Atezolizumab	Immune checkpoint inhibitor	Binds to PD-L1 allowing T cells to kill tumor cells	Intravenous infusion	NCT03125902	34226684
TRIPLE-NEGATIVE	Eribulin mesylate	Microtubule inhibitor	Inhibits growth of microtubules preventing cancer cell division	Intravenous infusion	NCT00965523	29604436
HER2+	Trastuzumab deruxtecan	Antibody-drug conjugate	Attaches to HER2 protein and delivers fatal chemicals into breast cancer cells	Intravenous infusion	NCT04420598	33961795
HER2+	Lapatinib + capecitabine	Kinase inhibitor	Binds to HER2 receptors to prevent tyrosine kinase activity	Orally	NCT00967031	34226684
HER2+	Margetuximab	Monoclonal Antibody	Bind HER2 receptors to prevent tumor cell growth	Intravenous infusion	NCT02492711	33480963
HER2+	Neratinib + capecitabine	Kinase inhibitor	Binds to HER receptors preventing cell proliferation	Orally	NCT00741260	34226684
HER2+	Tucatinib + Trastuzumab	Tyrosine kinase inhibitor	Binding to HER2 receptors decreasing cell proliferation	Orally	NCT02614794	33250512
TRIPLE-NEGATIVE	Olaparib	PARP inhibitor	Inhibition of PARP activity prevents cell oversight of DNA damage	Orally	NCT03286842	34226684
LUMINAL	Palbociclib + Fulvestrant	CDK4/6 inhibitor	Inhibits cell cycle activity preventing cell growth	Orally	NCT02491983	34226684
HER2+	Pyrotinib + capecitabine	Kinase Inhibitor	Binds to HER1, HER2, and HER4 to prevent cell proliferation	Orally	NCT03691051	34226684
LUMINAL	Ribociclib + letrozole	CDK4/6 inhibitor	Inhibits cell cycle proteins to slow progression of cancer	Orally	NCT03096847	34226684
TRIPLE-NEGATIVE	Sacituzumab govitecan	Antibody-drug conjugate	Targets and kills cancer cells	Intravenous infusion	NCT02574455	32529410
TRIPLE-NEGATIVE	Talazoparib	PARP inhibitor	Inhibition of PARP activity prevents cell oversight of DNA damage	Orally	NCT02401347	34226684
LUMINAL	Tamoxifen	Selective estrogen receptor modulator	Binds to hormone receptors to prevent cancer growth	Orally	NCT00002579	33736709
LUMINAL	Toremifene	Selective estrogen receptor modulator	Binds to hormone receptors to prevent cancer growth	Orally	NCT00044291	29978332
HER2+	Trastuzumab emtansine	Antibody-drug conjugate	Inhibition of HER2 receptor through binding	Intravenous infusion	NCT01702571	32881421
HER2+	Trastuzumab + Pertuzumab	Monoclonal antibody	Inhibits receptor dimerization of HER2-dependent cells by binding to its surface, preventing a response from the immune system	Intravenous infusion	NCT02131064	34226684

Newer treatments being studied focus on targeted therapies that interact with the hormone receptors expressed by the cancer cells. This includes the androgen receptor, which is typically neglected when considering treatment strategies[3]. It is frequently expressed in luminal type cancers and could make a valuable target when the cancer is resistant to therapies that target other receptors. Other new therapy options include inhibitors and trastuzumab-conjugated nanoparticles [Table 2], which could simultaneously deliver docetaxel and siRNA against HER2 to treat HER2-positive cancer[6]. Studies on triple-negative breast cancer are developing an acid-based small molecule nitrogen compound that is transported to the cell via large amino acid transporter type 1 (LAT1)[7]. These new therapeutic strategies may circumvent the drawbacks of current treatments and provide an even better prognosis for patients in the future.

Table 2. Emerging treatments for breast cancer brain metastasis

Type of breast cancer	Treatments	Treatment class	Mechanism of action	Route of administration	Clinical trial	PMID
HER2+	Afatinib	Tyrosine kinase inhibitor	Inhibits kinase activity of HER2 receptors	Oral	NCT00425854	32030570
TRIPLE-NEGATIVE	Atezolizumab and stereotactic radiation	CDK4/6 Inhibitor	Improve body's immune response to cancer cells	Orally and radiation	NCT03483012	33250512
HER2+	ANG4043	Peptide antibody conjugate	Targets and reduces size of HER2+ tumors	Intravenous infusion	N/A	25492620
LUMINAL	Bevacizumab	VEGF inhibitor	Inhibit formation of tumor cells	Intravitreal injection	NCT01190345	33552547
TRIPLE-NEGATIVE	Bicalutamide	Anti-AR antagonist	Blocks androgen receptors to prevent cell growth	Oral	NCT00468715	21633166
LUMINAL	Buparlisib	P13k inhibitor	Inhibits division of tumor cells by blocking kinase activity	Oral	NCT01790932	31552290
HER2+	GDC-0084 + trastuzumab	PI3K inhibitor	Inhibits division of tumor cells by blocking kinase activity	Orally and intravenously	NCT03765983	33250512
TRIPLE-NEGATIVE	Pembrolizumab and stereotactic radiation	Monoclonal antibody	Stimulate immune system to kill cancer cells	Intravenous infusion and radiation	NCT03449238	33250512
HER2+	Temozolomide and irinotecan	Alkylating agent	Destroys tumor cell DNA	Orally and intravenously	NCT00617539	31172405
TRIPLE-NEGATIVE	Veliparib and cisplatin	PARP inhibitor	Prevent DNA repair mechanisms in cancer cells	Orally and intravenously	NCT02595905	33250512
TRIPLE-NEGATIVE	QBS10072S	LAT1 small molecule chemotherapeutic	Targets LAT1 to suppress tumor growth	Intravenous	NCT04430842	34635566

BREAST CANCER BRAIN METASTASIS FORMATION

To properly understand the mechanisms of current and new therapeutic strategies for breast cancer that has metastasized to the brain, it is important to first understand the process of disease progression. Importantly, malignant breast tumors have a multitude of cell phenotypes, including cancer stem cells and cells that are multipotent, highly proliferating, and potentially multi-drug resistant. As the disease advances, malignant cells invade tissue through the basement membrane. They then intravasate into the blood vessels of the breast tissue as part of the epithelial-to-mesenchymal transition (EMT) [Figure 1]. These cells are carried throughout the circulatory system, eventually reaching other tissues of the body. The EMT gives cells stem-cell-like properties, which are known to contribute to brain metastasis and progression. Once these cells reach the central nervous system, they break through the blood-brain barrier and continue to circulate within the brain. These malignant cells eventually begin to proliferate and create new tumors within or around the brain[8]. As the tumors grow, there is increased pressure on the brain, which may lead to physical symptoms, including personality changes, memory loss, or seizures.

Some genes are associated with an increased risk of breast cancer and possibly point to how aggressive the disease may be. BRCA1 and BRCA2 gene mutations have been found to greatly increase the chance of a patient developing breast cancer by the age of 70[9]. Recently, the glycoprotein progranulin (PGRN) has been found to be a potential biomarker for increased risk of breast cancer development, simultaneously making a potential therapeutic target. PGRN activates cell signaling cascades that assist in the survival and proliferation of cancer cells and can be induced by estrogen[10].

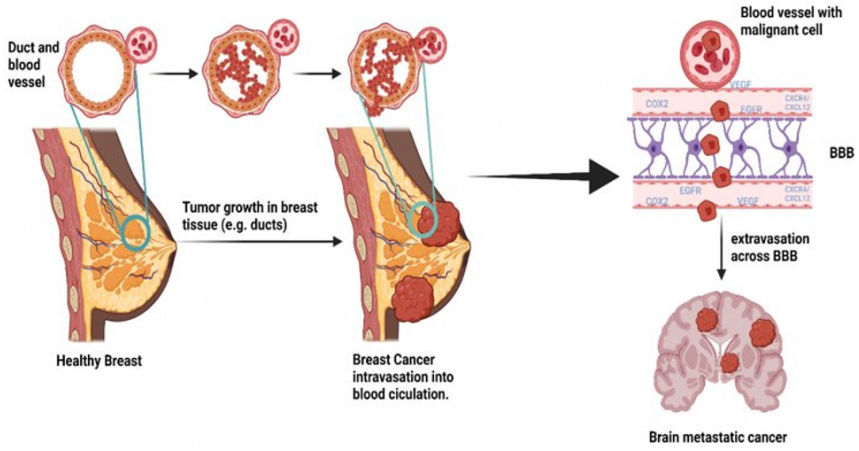

Figure 1. Pathway of disease progression of breast cancer leading to brain metastasis. (Image created in BioRender.com)

CURRENT TREATMENT METHODS

HER2-positive

Overexpression or amplification of human epidermal growth factor receptor 2 (HER2) is evident in about 15%-20% of breast cancer patients[11]. Normal amounts of HER2 protein receptors control the growth, division, and repair of healthy breast cells. However, elevated levels of this protein are indicative of rapid growth and excess proliferation of those cells, compared to those that are HER2-negative. The HER2-positive phenotype is regarded as an extremely aggressive form of breast cancer and is associated with poor prognosis, but it also implies that these cancers are much more susceptible to immunotherapy or other drug therapy that targets the HER2 receptor[12].

The emerging class of antibody-drug conjugates (ADCs) has altered treatment regimens for HER-2-positive metastatic breast cancer. This class of anti-tumor drugs combines the therapeutic advantages of monoclonal antibodies and cytotoxic drugs via linkers. Trastuzumab is a HER2-targeted humanized monoclonal antibody that binds to the extracellular domain of the protein [Table 1]. It was originally approved as a first-line therapy by Slamon *et al.* in a randomized study of patients who received either standard chemotherapy alone or a combination of chemotherapy with trastuzumab[11]. Deruxtecan is an ADC that contains trastuzumab as the anti-HER2 monoclonal antibody conjugated to an exatecan-derivative topoisomerase I inhibitor. ADC therapy of Trastuzumab deruxtecan was shown in clinical trials to have 60% overall response and up to 97% disease control in patients[12]. In a phase 3, multicenter, randomized trial (n = 524) comparing the efficacy and safety of trastuzumab deruxtecan against trastuzumab emtansine in patients with HER2-positive metastatic breast cancer, the authors concluded that the risk of disease progression or death was lower for patients who received trastuzumab deruxtecan (PFS = 75.8%, CI = 95%) than trastuzumab emtansine (PFS = 34.1%, CI = 95%)[13]. Subsequent to the results of this clinical trial, trastuzumab deruxtecan has become the new standard of care treatment for HER2-positive metastatic breast cancer[14]. Since these novel developments, targeting HER2 receptors with trastuzumab concomitantly or sequentially with neoadjuvant chemotherapy has radically improved the prognosis of HER2-positive breast cancers and become the cornerstone of treatment for metastatic breast cancer[15].

Once HER2 is dimerized by other epidermal growth factor receptor (EFGR) proteins, tyrosine kinase residues are phosphorylated, which in turn activates a host of downstream cascades that is responsible for cell growth, proliferation, and healing[16]. Trastuzumab can circumvent this dimerization by binding to the juxta-membrane domain of HER2, reducing HER2 signaling [Figure 2].

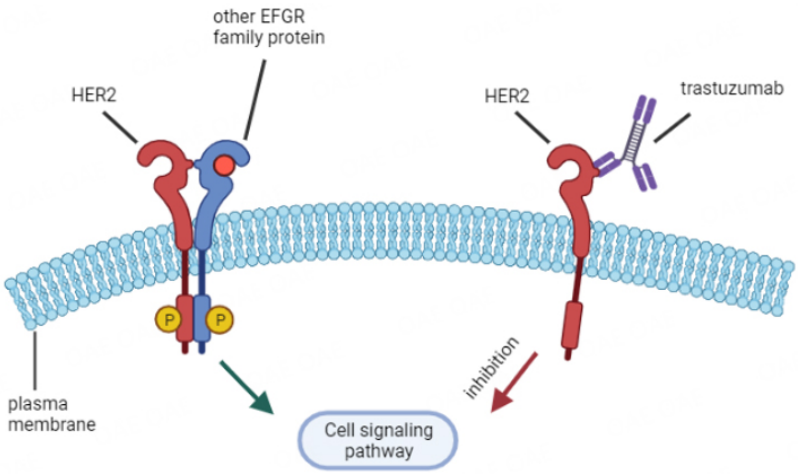

Figure 2. HER2 signaling pathway is activated by dimerization of HER2 to another protein. Trastuzumab targets the binding domain of HER2 and inhibits dimerization, thus blocking the cell signaling pathway. (Image created in BioRender.com)

The combination of trastuzumab with chemotherapy has a few drawbacks despite significantly improving prognosis and long-term survival. A follow-up study of patients enrolled in a trastuzumab adjuvant trial 11 years after treatment showed that > 1 year of trastuzumab treatment had no additional benefit, and about 25% of patients developed a recurrence[17]. Resistance to trastuzumab has also been observed; about 65% of patients with HER2-positive breast cancer that received trastuzumab initially did not respond, and about 70% of patients a year after initial treatment observed further progression of cancer. However, trastuzumab still remains the primary treatment option for HER2-positive breast cancer due to its established efficacy and safety[18].

Recent second-generation studies led to the approval of novel HER2 targeting agents such as pertuzumab, tyrosine kinase inhibitors including lapatinib and tucatinib, and second-generation antibody chemotherapy conjugates such as ado-trastuzumab emtansine (T-DM1) [Table 1][15,19]. Current studies are also looking to reduce resistance to HER2 targeting agents by approaching parts of the downstream pathways. A potential improvement may come from inhibiting phosphoinositide 3-kinase (PI3-K)/enzyme Ak transforming factor (Akt), a signaling pathway crucial for cell survival[20]. The pathophysiological pathways of the HER2 cascade have still been poorly mapped out; thus, more research is needed to further improve treatment by either combining different HER2 targeting agents or decreasing cell resistance to HER2 targeting antibodies. Understanding the mechanism of trastuzumab and its interactions with HER2 proteins on cancer cells in primary tumors is important to understand how metastatic tumors may interact with the drug. This makes the therapy a crucial part of combating metastatic breast cancer, especially when the cancer is not sufficiently responsive to chemotherapy alone due to resistance or inability to cross the blood-brain barrier.

Triple-negative

Triple-negative breast cancer (TNBC) is classified as a subtype of cancer that does not express estrogen receptors (ER), progesterone receptors (PR), or HER2 receptors[21]. Due to the cancer cells' unique phenotype, treatments that would usually work on breast cancer cells that express these receptors, such as endocrine or targeted therapies, are not effective against TNBC or metastasized tumors stemming from this type of cancer. While TNBC lacks a standardized care approach, current methods mainly consist of neoadjuvant chemotherapy, which is a preliminary treatment followed by surgery[22,23]. In fact, numerous literatures have suggested that neoadjuvant chemotherapy enhances remission of TNBC significantly more than cancers positive for hormone receptors. Common chemotherapies include anthracycline, alkylating

agents, taxanes, fluorouracil, and cyclophosphamide[21,23]. While chemotherapy drugs result in a better prognosis for TNBC patients[24], the lack of a standardized drug selection and limited options illuminates the importance of either optimizing chemotherapy regimens or researching other treatment modalities.

A small number of targeted molecular therapies have been approved, most notably poly ADP-ribose polymerase enzymes (PARP) inhibitors [Table 2], such as olaparib and veliparib[22]. PARPs are a family of enzymes that play a direct role in base excision repair, genome stability, and maintenance of the cell cycle[22,25]. PARP inhibitors have been used to target mutated breast cancer genes (BRCA), including BRCA1 and BRCA2. The BRCA genes are inherent tumor suppressors and are required for homologous recombination in cells. Mutations in these genes increase the risk of developing breast cancer by up to 70%[9]. However, the inability to perform homologous recombination in BRCA mutated cells creates a synthetic lethality and cells become hypersensitive to PARP inhibitors. Results in a study by Ding *et al.* showed that olaparib efficiently kills BRCA2-deficient cells via selective induction of synthetic lethality[9]. Some studies that examined the efficacy of PARP inhibitors have conflicting results, with some showing promising clinical benefits[26] while others did not find a significant difference between TNBC patients with and without a BRCA1/2 mutation. There is also potential for the development of drug resistance via restoration of homologous recombination in cancerous cells[27]. Despite this, PARP inhibition treatments have still been shown to give patients greater benefits in quality of life of patients compared to chemotherapy, but future preclinical trials are needed for ideal neoadjuvant results[22,25,28].

Other novel therapeutics that have been implemented in the clinic include antibody-drug conjugates such as sacituzumab govetican, and pembrolizumab. Similar to trastuzumab deruxtecan, sacituzumab govetican includes a topoisomerase I inhibitor conjugated to a monoclonal antibody. It is an anti-trophoblast cell surface monoclonal antibody, which is a marker for the antigen on the cell surface of trophoblasts commonly overexpressed in TNBC[29], and has become an effective single-agent therapeutic approach for metastatic TNBC. Pembrolizumab is an immune checkpoint inhibitor that has proven to be efficacious by inhibiting the programmed cell death protein 1 (PD-1)[30]. Immune checkpoint inhibitors (ICIs) are used to kick-start a patient's own immune system to respond and kill the cancerous cells that may normally evade immune detection by overcoming normal inhibitory pathways regulated by immune checkpoints. ICIs are given in order to remove the need for inhibitory pathway activation so the immune system can begin to identify cancerous cells[31]. In a phase 1 clinical trial, pembrolizumab was given intravenously and showed that the response rate was 18.5% out of the 27 patients evaluated[30] and has since become an effective monotherapy for TNBCs that have been previously treated.

Furthermore, the KEYNOTE-355 trial evaluating the overall survival among patients with pembrolizumab plus chemotherapy found this combination to result in significantly longer functional performance status (FPS) than chemotherapy for patients whose tumors expressed PD-L1 with a combined positive score of 10 or more[32]. An AICI that has had conflicting results in treating metastatic TNBC is atezolizumab, a PD-L1 inhibitor. Results from the IMpassion130 and IMpassion131 clinical trials evaluated atezolizumab added to nab-paclitaxel and paclitaxel, respectively. The results from the IMpassion130 clinical trial showed that atezolizumab plus nab-paclitaxel combination prolonged PFS by 1.2 months (95%CI: 0.62 to 0.92) than the placebo plus nab-paclitaxel[33]. However, results from a subsequent phase 3, double-blinded study, IMpassion131, showed that atezolizumab plus paclitaxel did not improve PFS or OS versus placebo plus paclitaxel[34]. One suggested explanation for the discrepancy in results is the use of different taxanes in the studies. Paclitaxel requires steroid premedication, which has been suggested to have a detrimental effect on ICI efficacy[35].

TNBC remains a highly aggressive subtype of breast cancer that generally has a poor prognosis for patients. Since the current methods still lack the effectiveness to provide patients with long-term benefits, it is crucial that alternative therapies are improved. Novel treatments like PARP inhibition have been introduced, but new targeting agents, improvement of standard treatment regimens, and a better immunological understanding of tumor progression of TNBC are needed to improve future patient outcomes.

Luminal A and luminal B

Luminal A and luminal B breast cancers encompass the two primary types of breast cancers that exhibit estrogen receptors. The key distinction between these groups lies in the proliferation rate of cancerous cells. This disparity is typically associated with the levels of the Ki-67 protein expressed, as luminal B cancers generally exhibit higher levels compared to luminal A cancers[1]. Another notable difference between these two subtypes is that luminal B breast cancer cells may express fewer estrogen receptors, resulting in reduced sensitivity to endocrine treatment. Furthermore, luminal B cancer can also produce more growth factors and can exhibit both progesterone-positive and progesterone-negative phenotypes. It also mildly expresses HER2 proteins and low levels of Ki-67 protein, which is a marker associated with cellular proliferation[36]. These factors generally contribute to a less favorable prognosis for luminal B cancer. Luminal A cancer is characterized by being both progesterone and estrogen positive, while also lacking any properties of HER2 or Ki-67 proteins. Consequently, luminal A cancers are considered lower grade than luminal B breast cancers, exhibiting a generally positive prognosis and lower risk of recurrence[18,37].

The identification and classification of luminal cancers typically involve gene-based assays and IHC-based markers[38]. Once diagnosed, the primary approach to treatment involves endocrine therapy. Due to the high presence of hormone receptors, luminal A often responds favorably to this type of treatment. Medications such as Tamoxifen or aromatase inhibitors act on estrogen receptors to specifically target cancer cells[39]. By inhibiting the estrogen receptors, the necessary components for cellular growth and proliferation are prevented, potentially leading to apoptosis. Estrogen inhibition may also prevent induction of PGRN, also prohibiting cancer cell proliferation[10].

By contrast, luminal B cancers tend to be more aggressive and usually require a combination of therapies. Most patients with luminal B type cancer, particularly those with low to no HER2 expression, receive endocrine therapies alongside some cytotoxic therapies. All patients with both luminal cancer types receive endocrine therapy because they exhibit hormonal receptor expression. Patients with HER2 expression in addition to luminal B cancer may receive endocrine therapy or cytotoxic therapy along with HER2 blockers to limit tumor growth[38].

THE BLOOD-BRAIN BARRIER

HER2+ and triple-negative subtypes have been studied to be two of the most aggressive types of breast cancer with the highest rates of brain metastasis[7,40]. An explanation as to why metastases originating from the most aggressive subtypes are unable to be treated efficiently may be due to a lack of specific targets to inhibit the cancer cell proliferation and the cancer cell's ability to penetrate the blood-brain barrier (BBB)[41]. The cancer cells' ability to cross the BBB allows the cells to circulate and eventually proliferate and differentiate in the brain, creating metastatic tumors. This may leave the BBB disrupted, which may allow for treatments to reach the tumor site. The BBB is normally selective in what is allowed to cross through, and its ability to do this can be attributed to the tight junctions on epithelial cells and selective transporters that specifically allow a specific size or nutrient to pass through. Astrocytes are glial cells that are specialized to maintain the BBB's integrity and have both tumor-killing and -promoting functions[41,42]. The tight junctions and epithelial layer may be broken by the cancer cells, which disrupts the barrier function of

preventing molecules from reaching the brain. Consequently, the function of the astrocytes may also be rendered ineffective in killing tumor cells. These properties, along with the possibility of cancer mutating with different properties from the primary tumor, are considerations that must be accounted for when creating a treatment strategy for breast cancer that has metastasized to the brain.

NEW DIRECTIONS

When metastasizing from the primary tumor, breast cancer cells have been shown to have the ability to undergo an epithelial-to-mesenchymal transition (EMT) that allows them to lose their epithelial cell phenotype. Specifically, tight junctions, apical-basal polarity, and the cytoskeleton allow for a more motile and invasive phenotype that becomes resistant to treatment[40,43,44]. Studies have shown that pSTAT3+ reactive astrocytes block CD8+ cytotoxic T cells from cancer cells and upregulate programmed cell death 1 (PDL-1) and vascular endothelial growth factor A (VEGF-A), as well as survival genes in cancer cells such as protocadherin 7 (PCDH7)[41]. Metastatic cancer cells in the brain also produce matrix metalloproteinases (MMPs) that degrade type IV collagen, the main vascular component of the basement membrane, thus enhancing the access points through the BBB [Figure 3][45]. Interestingly, the BBB is not disrupted in patients who have the HER2+ subtype; disruption is mostly seen in patients with triple-negative and basal-type cancers[42]. Treatments that inhibit the angiogenesis and survival processes of cancer cells are currently being studied to discover potent inhibitors for the pathways, as well as the transporters that carry these drugs through an intact BBB.

HER2-positive

Current breast cancer brain metastasis treatments focus on the HER2, VEGF, and EGFR P13K/Akt/mTOR pathways[43]. HER2+ treatment includes monoclonal antibodies inhibiting the human epidermal receptor (HER), including trastuzumab and pertuzumab. Inhibiting HER reduces the EGFR signaling pathway that contributes to the proliferation of cancer cells in the brain[45]. Trastuzumab and pertuzumab have shown promising results. However, these antibodies are too large to penetrate the intact BBB, therefore decreasing the inhibitory effectiveness of this treatment and should be used concurrently with chemotherapy to improve the survival rate of patients[45,46]. To bypass the intact BBB, Venishetty *et al.* focused their drug delivery on the choline transporter (CT), a transporter necessary for the acetylcholine pathway that has a moderate affinity to choline and has shown its effectiveness in drug delivery[47]. Previous studies have shown that the vascular endothelial growth factor (VEGF) pathway may contribute to enhancing brain metastasis by inducing the P13K/AkT/mTOR and STAT3 pathway allowing for transendothelial migration of tumor cells[43]. As a result, antibodies neutralizing the vascular endothelial growth factor receptor (VEGFR) have been shown effective in inhibiting the phosphorylation of vascular endothelial growth factor A, VEGF-A, and the proliferation and migration of cancer cells through the BBB[44,48].

Recent literature has shown that small molecule tyrosine kinases inhibitors (TKIs) have strong therapeutic promise for metastases [Table 2][49]. Unfortunately, some TKIs have off-target effects leading to frequent cases of diarrhea and skin toxicity[50]. Tucatinib is extremely selective for the HER2 receptor and only minimally inhibits other HER family receptors[3]. In the HER2CLIMB study, which consisted of a randomized, double-blinded, placebo-controlled trial of patients (n = 612) with locally advanced or metastatic HER2+ breast cancer, clinicians tested the efficacy of tucatinib combined with a trastuzumab and capecitabine regimen with respect to overall survival (OS) and progression-free survival (PFS). The clinicians report the median OS was 24.7 months for the tucatinib-included group versus 19.2 months for the control group (95%CI: P = 0.004). Half of the HER2CLIMB patients displayed brain metastases, significantly and accurately reflecting the real-world population, wherein approximately 50% of HER2+ metastatic breast cancer patients will develop brain metastases[50].

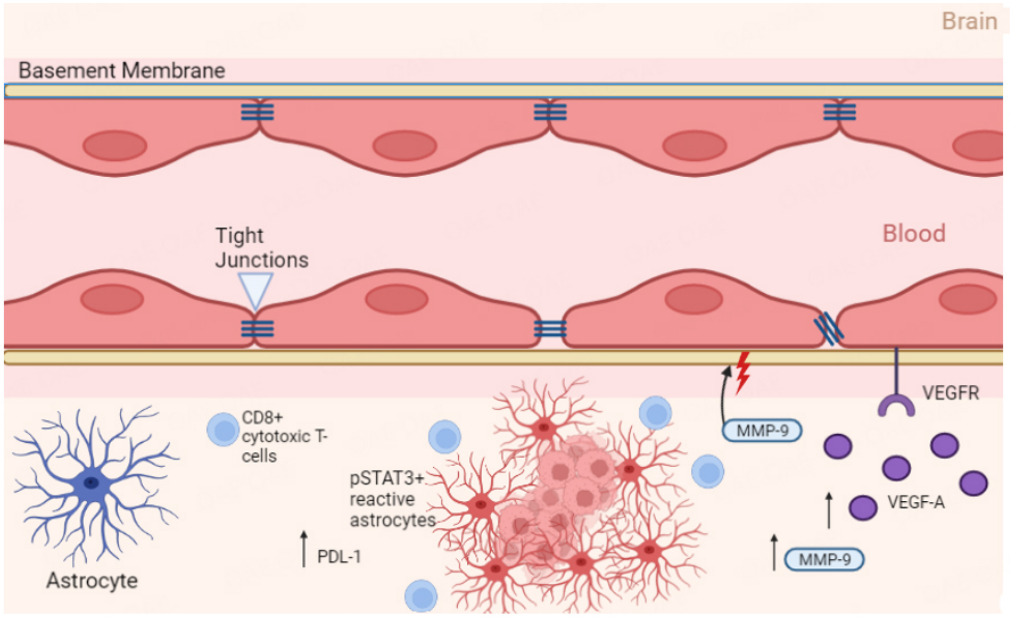

Figure 3. pSTAT3+ reactive astrocytes block CD8+ cytotoxic T cells from cancer cells and upregulate programmed cell death 1 (PDL-1) and vascular endothelial growth factor A (VEGF-A)[42]. Brain cancer cells also produce matrix metalloproteinases (MMPs) that degrade type IV collagen, the main vascular component of the basement membrane, thus enhancing the access points through the BBB[46]. The BBB is not disrupted in patients who have the HER2+ subtype. (Image created in BioRender.com)

Current treatment methods for advanced and metastatic HER2+ breast cancer include two HER2-targeting antibodies and a taxane. This standard regimen has many limitations, such as cost and lengthy infusion times. In response to these limitations, Ngamcherdtrakul *et al.* presented a novel therapeutic based on trastuzumab-conjugated nanoparticles for the simultaneous delivery of docetaxel and siRNA against HER2 (siHER2), T-siHER2-NP(DTX)[6]. The authors found that these optimized nanoparticles performed better than their free drug counterpart when used at the same dose. Additionally, when combined with microbubble-assisted focused ultrasound, T-siHER2-NP(DTX) had successful treatment outcomes in breast tumors residing in the brains of mice as opposed to T-siHER2-NP(DTX) alone ($P < 0.05$)[6].

Triple-negative

Triple-negative breast cancers (TNBCs) have the highest rate of brain metastasis and are the most aggressive type of cancer that has limited treatment options due to its phenotypic characteristic of not expressing any hormone receptors. Current treatments for this type of cancer are tailored around chemotherapy which targets high expressions of proteins that contribute to the high proliferation of these cancer cells.

Large amino acid transporter type 1 (LAT1) is upregulated in TNBCs and provides amino acids to tumor cells for rapid proliferation. In their study, Deng *et al.* have developed an amino acid-based small molecule nitrogen compound (QBS10072S) that is transported by LAT1 across the blood-brain barrier (BBB)[51]. QBS10072S is a non-cleavable amino acid molecule that is recognized by LAT1 which is highly expressed in TNBC cells. When compared to melphalan, a related nitrogen mustard compound, the authors report that QBS10072S showed a 25-fold higher specificity for LAT1, and lower cytotoxicity with respect to normal cells. After an immunohistochemistry analysis, the authors report that LAT1 is highly expressed in TNBC brain metastases[7]. Importantly, LAT1 levels were substantially higher in the brain metastases than in surrounding brain tissue, making QBS10072S a potential therapeutic for TNBC brain metastasis. Treatment with QBS10072S, administered either intravenously or intraperitoneally, resulted in a significant reduction

in intracranial brain tumor growth ($P < 0.0001$) and an increase in OS ($P < 0.002$, $P < 0.04$ respectfully) in comparison to vehicle groups. Ultimately, the novel therapeutic QBS10072S is highly selective to TNBCs and has been shown to significantly slow TNBC's proliferation rate[51]. Although this study shows a more targeted treatment for TNBCs, it should be mentioned that the effectiveness of the drug relies on the LAT1 expression in TNBCs, and expression levels may differ from patient to patient.

Luminal A and luminal B

Brain metastases are a common byproduct of aggressive breast cancers such as HER2+ and triple-negative. However, a study conducted by Oehrlich *et al.* suggests that patients with luminal breast cancer have a lower risk of developing brain metastases[52]. Luminal A tumors are characterized to display high expressions of estrogen-related genes and low expression of proliferation genes, while luminal B tumors display lower expression of estrogen and progesterone receptor genes and high expression of proliferation, cell cycle genes, and growth factor receptor genes[36]. Both luminal A and luminal B cancer are positive for estrogen receptor (ER+), but luminal B has lower expression in comparison to luminal A. Luminal B also has higher proliferation rates, which contributes to its aggressive phenotype[6,51].

Treatments targeting luminal B tumors focus on the proliferating phenotype of these cells due to the responsiveness they have against hormones and chemotherapy[3,6,51]. Inhibition of the P13K/AkT/mTOR and ERK pathways reduces the survival, angiogenesis, and metastasis of these tumor cells[36]. Due to luminal A and B's high estrogen receptor expression, drugs like Tamoxifen are used to bind to the receptors and prevent cancer cells from developing resistance to the innate anti-cancer mechanisms[3]. Due to the potential of developing tamoxifen resistance and current cytotoxic therapies, insulin-like growth factor-1 receptor (IGF-1R) is becoming a potential target for luminal cancer treatment. This pathway has been found to have a great effect on limiting tumor proliferation. Most resistance to Tamoxifen derives from the communication between ER and IGF-1R receptors in the tumor cells[53]. The PR loss is associated with the IGF-1R in Luminal B Cancer cells. The IGF-1R inhibitor BMS-536924 significantly suppressed cancer growth *in vitro* in various cell lines, including MAC-7 Tam-R cells, which have tamoxifen resistance. IGF-1R inhibitors may become a novel therapeutic strategy, but even identifying the presence of the receptor has the potential to become a significant prognostic factor in diagnosis[54].

In a 2018 study, Hwang *et al.* evaluated the efficacy of tamoxifen as a therapeutic for luminal A subtype breast cancer [Table 1][55]. The authors report that luminal A subtype showed a higher survival rate compared to TNBC ($P = 0.009$)[55]. Androgen receptor (AR), while not commonly noted, is frequently expressed in luminal A and B subtypes of breast cancer and thus may be a potential therapeutic target. In their study, Bergen *et al.* evaluated AR expression within the tumor-cell nucleus by performing immunohistochemical staining of AR on brain metastases samples from patients with breast cancer ($n = 57$)[56]. They report that AR expression ≥ 1% was evident in 35.1% of samples and the median AR expression rate was 10%. AR expression in ≥ 1% of tumor cells was observed in 52.4% of brain metastases of the luminal/HER2-negative subtype and 23.1% of the luminal/HER2-positive subtype. This suggests that there is potential for the development of future therapeutics that target AR expression in luminal type cancers.

Nanotechnology

A prevailing obstacle in the development of therapeutics targeting brain metastases is the inability of drugs to cross the BBB. As such, any potential drug targeting brain metastases must be BBB permeable. One of the newest strategies for the treatment of all types of metastases originating from breast cancer is using a nano-sized drug delivery system. Some newly investigated nano-drugs include: the PEGylated nano-liposomal doxorubicin; the non-pegylated liposomal doxorubicin; albumin-bound paclitaxel; and a polymeric

nanoparticle (NP) micelle formulation of paclitaxel[57]. The nano-liposomal doxorubicin was also the first FDA-approved nano-drug authorized for clinical use. Another strategy includes co-delivery of drugs, such as the synthesis of 5-FU and paclitaxel (PTX)-loaded KLA-modified liposomes for the improved treatment of triple-negative breast cancer[58].

An in vitro and animal model study by Sakhi *et al*. demonstrated that the developed PLGA-paclitaxel nano-formulations conjugated with trastuzumab had favorable physiochemical characteristics, surface morphology, sustained release kinetics, and enhanced targeting against HER2+ breast cancer cell lines[59]. Another study by Elhabak *et al*. modified single step nanoprecipitation method using a long circulating nanocarrier comprising trastuzumab (TZB) surface modified polylactic-*co*-glycolic acid (PLGA) nanoparticles (NPs) *co*-encapsulating magnolol (Mag) and gold nanoparticles (GNPs) demonstrated that optimized NPs were able to boost Mag cytotoxicity on breast cancer cells while providing a selective multifunctional therapy with an added photothermal effect[60].

Nanoparticles are becoming an increasingly popular area of interest as they have the potential to penetrate the BBB. Due to the nature of the disease, the BBB may no longer be fully intact once the cancerous cells pierce it, and there will be an increase in macrophage recruitment to the area. It is possible that nanoparticle-laden monocytes or macrophages may make a perfect vehicle to cross the BBB and reach the metastatic brain lesions[61].

CONCLUSION

Metastatic breast cancer can be a harrowing diagnosis for many patients, especially when the cancer has mutated or gained resistance to certain therapeutic strategies. It can be especially traumatic when these metastases reach the brain, which is a vitally important and delicate organ. Current treatments typically involve a combination of at least two therapies, depending on the histology and expression types of the cancer. Many of these therapies have severe toxicities and side effects that affect the patient's quality of life. Novel therapeutics aim to take advantage of previously unfamiliar targets with the goal of overcoming resistance that exists with current mechanisms. Although these new approaches have been found to have their own side effects and drawbacks, they are still considered significant advances in the field. They promise improvement in the prognoses of patients with breast cancer that has metastasized to the brain, from higher quality of life to increased remission rates and a greater chance of long-term survival.

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Authors' contributions

Made substantial contributions: Klaas E, Sung E, Azizi E, Martinez M, Barpujari A, Roberts J, Lucke-Wold B

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REFERENCES

1. DePolo J. Molecular subtypes of breast cancer. Available from: https://www.breastcancer.org/types/molecular-subtypes [Last accessed on 13 June 2023].
2. Breast cancer molecular subtypes |CTCA| city of hope. Available from: https://www.cancercenter.com/cancer-types/breast-cancer/types/breast-cancer-molecular-types [Last accessed on 13 June 2023].
3. Kulukian A, Lee P, Taylor J, et al. Preclinical activity of HER2-selective tyrosine kinase inhibitor tucatinib as a single agent or in combination with trastuzumab or docetaxel in solid tumor models. *Mol Cancer Ther* 2020;19:976-87. DOI
4. Most common molecular subtypes of breast cancer |CTCA|. Available from: https://www.cancercenter.com/cancer-types/breast-cancer/types/breast-cancer-molecular-types [Last accessed on 13 June 2023].
5. Inic Z, Zegarac M, Inic M, et al. Difference between luminal a and luminal b subtypes according to Ki-67, tumor size, and progesterone receptor negativity providing prognostic information. *Clin Med Insights Oncol* 2014;8:107-11. DOI PubMed PMC
6. Ngamcherdtrakul W, Bejan DS, Cruz-Muñoz W, et al. Targeted nanoparticle for co-delivery of HER2 siRNA and a taxane to mirror the standard treatment of HER2+ breast cancer: efficacy in breast tumor and brain metastasis. *Small* 2022;18:e2107550. DOI PubMed PMC
7. Nam BH, Kim SY, Han HS, et al. Breast cancer subtypes and survival in patients with brain metastases. *Breast Cancer Res* 2008;10:R20. DOI PubMed PMC
8. Wang Y, Ye F, Liang Y, Yang Q. Breast cancer brain metastasis: insight into molecular mechanisms and therapeutic strategies. *Br J Cancer* 2021;125:1056-67. DOI PubMed PMC
9. Ding X, Ray Chaudhuri A, Callen E, et al. Synthetic viability by BRCA2 and PARP1/ARTD1 deficiencies. *Nat Commun* 2016;7:12425. DOI PubMed PMC
10. Purrahman D, Mahmoudian-Sani MR, Saki N, Wojdasiewicz P, Kurkowska-Jastrzębska I, Poniatowski ŁA. Involvement of progranulin (PGRN) in the pathogenesis and prognosis of breast cancer. *Cytokine* 2022;151:155803. DOI PubMed
11. Slamon DJ, Clark GM, Wong SG, Levin WJ, Ullrich A, McGuire WL. Human breast cancer: correlation of relapse and survival with amplification of the HER-2/neu oncogene. *Science* 1987;235:177-82. DOI PubMed
12. Andrikopoulou A, Zografos E, Liontos M, Koutsoukos K, Dimopoulos MA, Zagouri F. Trastuzumab deruxtecan (DS-8201a): the latest research and advances in breast cancer. *Clin Breast Cancer* 2021;21:e212-9. DOI PubMed
13. Cortés J, Kim SB, Chung WP, et al. Trastuzumab deruxtecan versus trastuzumab emtansine for breast cancer. *N Engl J Med* 2022;386:1143-54. DOI
14. Ji C, Li F, Yuan Y, et al. Novel anti-HER2 antibody-drug conjugates versus T-DM1 for HER2-positive metastatic breast cancer after tyrosine kinase inhibitors treatment. *Oncologist* 2023;28:e859-66. DOI
15. Bredin P, Walshe JM, Denduluri N. Systemic therapy for metastatic HER2-positive breast cancer. *Semin Oncol* 2020;47:259-69. DOI PubMed
16. Vu T, Claret FX. Trastuzumab: updated mechanisms of action and resistance in breast cancer. *Front Oncol* 2012;2:62. DOI PubMed PMC
17. Cameron D, Piccart-Gebhart MJ, Gelber RD, et al. 11 years' follow-up of trastuzumab after adjuvant chemotherapy in HER2-positive early breast cancer: final analysis of the HERceptin Adjuvant (HERA) trial. *Lancet* 2017;389:1195-205. DOI PubMed PMC
18. Maximiano S, Magalhães P, Guerreiro MP, Morgado M. Trastuzumab in the treatment of breast cancer. *BioDrugs* 2016;30:75-86. DOI PubMed
19. Figueroa-Magalhães MC, Jelovac D, Connolly R, Wolff AC. Treatment of HER2-positive breast cancer. *Breast* 2014;23:128-36. DOI PubMed PMC
20. Hudis CA. Trastuzumab-mechanism of action and use in clinical practice. *N Engl J Med* 2007;357:39-51. DOI PubMed
21. Yin L, Duan JJ, Bian XW, Yu SC. Triple-negative breast cancer molecular subtyping and treatment progress. *Breast Cancer Res* 2020;22:61. DOI PubMed PMC
22. Lyons TG. Targeted therapies for triple-negative breast cancer. *Curr Treat Options Oncol* 2019;20:82. DOI PubMed
23. Won KA, Spruck C. Triple-negative breast cancer therapy: current and future perspectives (review). *Int J Oncol* 2020;57:1245-61. DOI PubMed PMC
24. Plevritis SK, Munoz D, Kurian AW, et al. Association of screening and treatment with breast cancer mortality by molecular subtype in US women, 2000-2012. *JAMA* 2018;319:154-64. DOI PubMed PMC
25. Kumar P, Aggarwal R. An overview of triple-negative breast cancer. *Arch Gynecol Obstet* 2016;293:247-69. DOI PubMed
26. O'Shaughnessy J, Osborne C, Pippen JE, et al. Iniparib plus chemotherapy in metastatic triple-negative breast cancer. *N Engl J Med* 2011;364:205-14. DOI

27. Kim DS, Camacho CV, Kraus WL. Alternate therapeutic pathways for PARP inhibitors and potential mechanisms of resistance. *Exp Mol Med* 2021;53:42-51. DOI PubMed PMC
28. Gelmon KA, Tischkowitz M, Mackay H, et al. Olaparib in patients with recurrent high-grade serous or poorly differentiated ovarian carcinoma or triple-negative breast cancer: a phase 2, multicentre, open-label, non-randomised study. *Lancet Oncol* 2011;12:852-61. DOI
29. Spring LM, Nakajima E, Hutchinson J, et al. Sacituzumab govitecan for metastatic triple-negative breast cancer: clinical overview and management of potential toxicities. *Oncologist* 2021;26:827-34. DOI PubMed PMC
30. Nanda R, Chow LQ, Dees EC, et al. Pembrolizumab in patients with advanced triple-negative breast cancer: phase Ib KEYNOTE-012 study. *J Clin Oncol* 2016;34:2460-7. DOI PubMed PMC
31. Thomas R, Al-Khadairi G, Decock J. Immune checkpoint inhibitors in triple negative breast cancer treatment: promising future prospects. *Front Oncol* 2020;10:600573. DOI PubMed PMC
32. Cortes J, Rugo HS, Cescon DW, et al. Pembrolizumab plus chemotherapy in advanced triple-negative breast cancer. *N Engl J Med* 2022;387:217-26. DOI
33. Schmid P, Adams S, Rugo HS, et al. Atezolizumab and nab-paclitaxel in advanced triple-negative breast cancer. *N Engl J Med* 2018;379:2108-21. DOI
34. Miles D, Gligorov J, André F, et al. LBA15 Primary results from IMpassion131, a double-blind placebo-controlled randomised phase III trial of first-line paclitaxel (PAC) ± atezolizumab (atezo) for unresectable locally advanced/metastatic triple-negative breast cancer (mTNBC). *Ann Oncol* 2020;31:S1147-8. DOI
35. Van Wambeke S, Gyawali B. Atezolizumab in metastatic triple-negative breast cancer-no contradiction in the eyes of a dispassionate observer. *JAMA Oncol* 2021;7:1285-6. DOI PubMed
36. Tran B, Bedard PL. Luminal-B breast cancer and novel therapeutic targets. *Breast Cancer Res* 2011;13:221. DOI PubMed PMC
37. Ignatiadis M, Sotiriou C. Luminal breast cancer: from biology to treatment. *Nat Rev Clin Oncol* 2013;10:494-506. DOI PubMed
38. Gao JJ, Swain SM. Luminal A breast cancer and molecular assays: a review. *Oncologist* 2018;23:556-65. DOI PubMed PMC
39. Treatment by cancer type. Available from: https://www.nccn.org/guidelines/category_1 [Last accessed on 13 June 2023].
40. Bailleux C, Eberst L, Bachelot T. Treatment strategies for breast cancer brain metastases. *Br J Cancer* 2021;124:142-55. DOI PubMed PMC
41. Hosonaga M, Saya H, Arima Y. Molecular and cellular mechanisms underlying brain metastasis of breast cancer. *Cancer Metastasis Rev* 2020;39:711-20. DOI PubMed PMC
42. Liang Y, Zhang H, Song X, Yang Q. Metastatic heterogeneity of breast cancer: molecular mechanism and potential therapeutic targets. *Semin Cancer Biol* 2020;60:14-27. DOI
43. Lah TT, Novak M, Breznik B. Brain malignancies: glioblastoma and brain metastases. *Semin Cancer Biol* 2020;60:262-73. DOI PubMed
44. Lamouille S, Xu J, Derynck R. Molecular mechanisms of epithelial-mesenchymal transition. *Nat Rev Mol Cell Biol* 2014;15:178-96. DOI PubMed PMC
45. Pedrosa RMSM, Mustafa DA, Soffietti R, Kros JM. Breast cancer brain metastasis: molecular mechanisms and directions for treatment. *Neuro Oncol* 2018;20:1439-49. DOI PubMed PMC
46. Shah N, Mohammad AS, Saralkar P, et al. Investigational chemotherapy and novel pharmacokinetic mechanisms for the treatment of breast cancer brain metastases. *Pharmacol Res* 2018;132:47-68. DOI PubMed PMC
47. Venishetty VK, Geldenhuys WJ, Terell-Hall TB, et al. Identification of novel agents for the treatment of brain metastases of breast cancer. *Curr Cancer Drug Targets* 2017;17:479-85. DOI PubMed PMC
48. Knier NN, Pellizzari S, Zhou J, Foster PJ, Parsyan A. Preclinical models of brain metastases in breast cancer. *Biomedicines* 2022;10:667. DOI PubMed PMC
49. Saura C, Oliveira M, Feng YH, et al. Neratinib plus capecitabine versus lapatinib plus capecitabine in HER2-positive metastatic breast cancer previously treated with ≥ 2 HER2-directed regimens: phase III NALA trial. *J Clin Oncol* 2020;38:3138-49. DOI
50. Curigliano G, Mueller V, Borges V, et al. Tucatinib versus placebo added to trastuzumab and capecitabine for patients with pretreated HER2+ metastatic breast cancer with and without brain metastases (HER2CLIMB): final overall survival analysis. *Ann Oncol* 2022;33:321-9. DOI
51. Deng J, Chernikova SB, Wang Y, et al. A novel brain-permeant chemotherapeutic agent for the treatment of brain metastasis in triple-negative breast cancer. *Mol Cancer Ther* 2021;20:2110-6. DOI PubMed PMC
52. Oehrlich NE, Spineli LM, Papendorf F, Park-Simon TW. Clinical outcome of brain metastases differs significantly among breast cancer subtypes. *Oncol Lett* 2017;14:194-200. DOI PubMed PMC
53. Peyrat JP, Louchez MM, Lefebvre J, et al. Plasma insulin-like growth factor-1 (IGF-1) concentrations in human breast cancer. *Eur J Cancer* 1993;29A:492-7. DOI
54. Law JH, Habibi G, Hu K, et al. Phosphorylated insulin-like growth factor-i/insulin receptor is present in all breast cancer subtypes and is related to poor survival. *Cancer Res* 2008;68:10238-46. DOI
55. Hwang KT, Kim EK, Jung SH, et al. Tamoxifen therapy improves overall survival in luminal A subtype of ductal carcinoma in situ: a study based on nationwide Korean Breast Cancer Registry database. *Breast Cancer Res Treat* 2018;169:311-22. DOI
56. Bergen ES, Berghoff AS, Steindl A, et al. Androgen receptor is expressed in breast cancer brain metastases. *Appl Immunohistochem Mol Morphol* 2021;29:728-33. DOI

57. Mercatali L, Vanni S, Miserocchi G, et al. The emerging role of cancer nanotechnology in the panorama of sarcoma. *Front Bioeng Biotechnol* 2022;10:953555. DOI PubMed PMC
58. Valencia-Lazcano AA, Hassan D, Pourmadadi M, et al. 5-fluorouracil nano-delivery systems as a cutting-edge for cancer therapy. *Eur J Med Chem* 2023;246:114995. DOI
59. Sakhi M, Khan A, Iqbal Z, et al. Design and characterization of paclitaxel-loaded polymeric nanoparticles decorated with trastuzumab for the effective treatment of breast cancer. *Front Pharmacol* 2022;13:855294. DOI PubMed PMC
60. Elhabak M, Osman R, Mohamed M, El-Borady OM, Awad GAS, Mortada N. Near IR responsive targeted integrated lipid polymer nanoconstruct for enhanced magnolol cytotoxicity in breast cancer. *Sci Rep* 2020;10:8771. DOI PubMed PMC
61. Choi MR, Bardhan R, Stanton-Maxey KJ, et al. Delivery of nanoparticles to brain metastases of breast cancer using a cellular Trojan horse. *Cancer Nanotechnol* 2012;3:47-54. DOI PubMed PMC