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Disrupting the metastatic cascade: nanoparticle-based innovations in antimetastatic therapy

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Antimetastatic therapy represents a transformative shift in oncology, addressing one of the greatest challenges in cancer care: the prevention and management of metastasis. By targeting the critical steps of the metastatic cascade and integrating them into multimodal treatment strategies, these therapies have the potential to greatly improve patient survival, enhance quality of life, and advance cancer care worldwide. Each stage of the metastatic cascade presents unique therapeutic vulnerabilities and opportunities for targeted intervention. Nanoparticle-based therapies have emerged as a promising frontier for disrupting the process of metastasis due to their ability to deliver therapeutic agents with high precision, reduce off-target effects of treatment, and overcome biological barriers. This review explores key targets suitable for antimetastatic therapy and recent advances in strategies using these modalities, highlighting their potential to improve treatment specificity and efficacy. By examining the integration of nanotechnology into antimetastatic therapy, we aim to underscore its transformative potential in combating metastatic cancer and improving patient outcomes.

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1. Metastatic cascade

To this day, secondary tumor foci – metastases – remain the leading cause of cancer-related morbidity and mortality.¹ To colonize distant sites, cancer cells have to undergo several fundamental events collectively termed “the metastatic cascade”, wherein such activation of invasion and metastasis is specifically recognized as a hallmark of cancer.² The process is initiated by the invasion of primary tumor cells into their local surroundings, followed by intravasation into the bloodstream or lymphatic system. Circulating tumor cells (CTCs) must then survive in blood circulation and lymphatic systems, extravasate, and proliferate in distant organs. The initial switch of the metastatic process triggering the tumor dissemination arises from a set of morphological, metabolic, and functional changes cancer cells undergo, collectively termed the epithelial–mesenchymal transition (EMT).³ This process is generally employed in cell development, during embryogenesis, and wound healing.⁴ However, in this case, it is hijacked by cancer cells, enabling them to lose epithelial properties and obtain certain mesenchymal features such as increased motility or

invasiveness, and resistance to anoikis.⁵ These changes are critical for successful cancer progression and play a key role at every stage of the metastatic cascade including invasion, intravasation, and extravasation. During EMT, epithelial cells lose cell–cell junctions, reorganize their cytoskeleton, and alter their gene expression patterns leading to a transition from polarized, cuboidal epithelial cells to mesenchymal spindle cells.

EMT is often triggered by microenvironment changes such as inflammation, hypoxia, and exposure to cytokines and growth factors.⁶ Many studies report “partial EMT” or cells residing in a spectrum of intermediate states between epithelial and mesenchymal phenotypes leading to a re-evaluation of how EMT is defined and understood in the context of cancer progression.^{7,8} Metastasis exhibits organ-specific distribution. The “seed and soil” hypothesis suggests that tumor spreading is guided by the cooperation of the malignant cells (“seeds”) and host organ environment (“soil”) and therefore, proceeds in a specific manner.⁹ Cancer stem cells (CSCs) possessing tumor initiation and phenotypic plasticity properties also contribute to the tumorigenic and malignant potential of tumors.¹⁰ In general, the metastatic progression can follow either a linear or parallel model.¹¹ While the linear model suggests a high genetic similarity in primary tumors and subsequently evolved metastases, in the parallel model the early disseminated tumor cells acquire metastatic capabilities independently of the primary tumor. Together these models underscore the complexity of the cancer metastasis process

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including the phenotypic and genetic diversity between primary and metastatic tumors.

1.1 Local invasion

At certain critical points, the tumor size and progression are severely limited by the amount of oxygen and nutrients supplied by blood vessels.¹² To access the distant sites, tumor cells must successfully invade the stromal and vasculature surroundings of their primary tumor. Establishing the tumor's own neovasculature beyond preexisting blood vessels is therefore a decisive step in the evolution of metastasis.¹³ Not only does it provide crucial nutrients and oxygen for the progression of the primary tumor, but it also offers a route for the dissemination of tumor cells into hematogenous circulation and to colonize distant anatomic sites. This transition is referred to as an "angiogenic switch".¹⁴ This component of tumor development is initiated by a disturbance of the equilibrium between pro- and antiangiogenic factors.¹⁵ While the prevalence of antiangiogenic aspects is associated with tumor dormancy, inclining the balance towards pro-angiogenic factors results in tumor angiogenesis and overall tumor proliferation. The angiogenic switch is triggered by a complex interplay of factors, including the release of pro-angiogenic molecules such as vascular endothelial growth factor (VEGF) by cancer cells, the protease degradation of the extracellular matrix and recruitment of endothelial cells to the tumor site. Once this switch is turned on, the formation of new vasculature begins and supplies the tumor with oxygen and nutrients, allowing it to grow and spread. Therefore, the angiogenic switch is a crucial therapeutic target and inhibiting angiogenesis can prevent further tumor development.¹⁶ Several anti-angiogenic drugs are currently in use, including bevacizumab (Avastin[®]) and sorafenib (Nexavar[®]).¹⁷ Bevacizumab is a monoclonal antibody that specifically targets VEGF and prevents it from binding to endothelial cell receptors and thereby inhibiting angiogenesis. Sorafenib is a kinase inhibitor that blocks angiogenesis-promoting signaling pathways by targeting several receptors involved in angiogenesis, including VEGF receptors, platelet-derived growth factor (PDGF) receptors, and Raf kinase. However, there are also challenges associated with anti-angiogenic therapy, including resistance and many side effects often limiting their use in certain patient populations.¹⁶

Another process also observed in primary tumors is lymphangiogenesis. In cancer, lymphangiogenesis can promote the spread of tumor cells to nearby lymph nodes and distant organ sites, a process known as lymphatic metastasis.¹⁸ Therefore, the presence of metastasis in lymph nodes often serves as an early prognostic indicator of metastatic dissemination and tumor invasiveness, particularly in carcinomas or melanomas.¹⁹ Additionally, changes in lymphatic endothelial cell (LEC) characteristics play a role in the formation of a premetastatic niche. A signaling pathway involving VEGF-C and PI3K α activates integrin $\alpha 4\beta 1$ on LECs, promoting expansion of lymphatic endothelium and enhancing the capture of metastatic cells expressing vascular cell adhesion molecule-1 (VCAM-1).²⁰ Furthermore, cancer-associated fibroblasts (CAFs)

producing high levels of periostin can compromise the lymphatic endothelial barrier by activating the integrin-FAK/Src-VE-cadherin pathway in LECs, therefore promoting lymph node metastasis.²¹ Integrin αIIB is also upregulated in LECs of tumor-draining lymph nodes, enhancing their ability to bind fibrinogen, which supports the adherence and survival of metastatic tumor cells.²²

1.2 Intravasation and survival

Successful intravasation of tumor cells provides a route for circulating tumor cells (CTCs) that are shed into the bloodstream by primary tumors and can be found in the peripheral blood of cancer patients.²³ CTCs can move either as individual cells or as clumps of cells that can survive circulatory stresses until they encounter microvessels in distant sites.²⁴ Nowadays, studying their molecular and functional characteristics represents an important area of cancer research of highly lethal cancers. CTCs are difficult to detect as they only represent a small fraction of the total cells in the blood.²⁵ CTC analysis can be used to identify specific mutations or biomarkers associated with drug resistance or other clinical outcomes and can help with overall treatment decisions.^{26,27} Moreover, by studying CTCs we can also gain insights into the mechanisms of cancer metastasis, providing opportunities for the development of new targeted therapies.

The transport and survival of CTCs is a complex process that involves several factors. CTCs need to evade the immune system,²⁸ withstand shear forces,²⁹ and adhere to blood vessel endothelium³⁰ in order to extravasate and form metastasis. Shear forces of the bloodstream cause huge mechanical stress on CTCs, leading to their rupture or deformation. Some CTCs can withstand such forces by altering their cell membrane proteins and cytoskeleton.³¹ Evading the immune system is possible by downregulating the expression of MHC class I molecules that are recognized by immune cells, reducing their visibility to the immune system.³² Additionally, CTCs can secrete factors that suppress immune cells, such as transforming growth factor beta (TGF- β).³³ During transport, CTCs also interact with other blood cells, especially platelets, which is mediated by the expression of integrins and selectins.³⁴ When platelets encounter CTCs, they can adhere to them and form complexes called circulating-tumor cell-platelet aggregates (CTC-PAs). CTC-PAs then travel through hematogenous circulation, providing CTCs with many benefits, such as forming a protective shield around them, shielding them from immune cells, and preventing their destruction. These adhesions enable cancer cells to evade natural killer cells due to the upregulation of the CD155 inhibitory checkpoint.^{35,36} Moreover, platelets can assist in the extravasation of CTCs into nearby tissues and promote adhesion to the endothelial cells that line blood vessels.^{37,38} Specifically, P-selectin responsible for the cell arrest at activated endothelium is being investigated as a potential nanotherapeutic target due to its high expression in tumor tissue compared to healthy tissue.³⁹ Furthermore, platelets promote the formation of small blood clots or microthrombi that create a favorable environment for establishing



new tumor sites. Understanding the precise mechanisms of these interactions as well as identifying selective pathways for inhibiting them, may translate into establishment of promising antimetastatic therapies. Neutrophil extracellular traps (NETs) have also emerged as major factors facilitating cancer metastasis. CTCs typically invade DNA- and protein-rich NETs, which generates protective particles that hide CTCs from immune responses, hematogenous and mechanical stresses, and facilitate CTC adhesion to endothelial cells.⁴⁰ In addition, components of NETs, including histones, proteases, and antimicrobial peptides, can degrade extracellular matrix (ECM) components, increasing the invasive ability of tumor cells.⁴¹ NETs may provide pro-tumorigenic pathways by sustaining tumor-induced inflammation conducive to metastatic colonization and growth.

Another mechanism involved in the survival of cancer cells in circulation is adhesive support. Loss of attachment to the extracellular matrix or neighboring cells often leads to anoikis, a type of programmed cell death. Resistance to anoikis is a key mechanism that allows survival of cancer cells since the bloodstream represents a hostile environment for CTCs with a lack of extracellular matrix.^{42,43} The resistance mechanisms involve alterations in gene expression or the formation of the above-mentioned CTC-PAs.⁴⁴ Research has shown that CTCs resisting anoikis are likely to successfully extravasate and establish new

tumors, subsequently evade immune surveillance, and resist chemotherapy and other treatments of cancer.⁴⁵ For this reason, targeting anoikis in cancer therapy may further improve the prevention of cancer metastasis.

1.3 Extravasation

Another challenge for tumor cells disseminated from primary tumors and surviving stresses in the bloodstream is their invasion through the endothelial vasculature into secondary tissues, a process known as transendothelial migration (TEM). The complex process of homing disseminated cancer cells into the secondary organs is still not fully understood. However, interactions between cancer cells and the target organ microenvironment, involving signaling molecules providing cues for CTCs to adhere, invade, and establish a metastasis, are crucial. Key steps and actions of successful extravasation include the adhesion of CTCs to the endothelial cells lining the blood vessels of target organs *via* surface adhesion molecules such as integrins and selectins, subsequent rolling of CTCs along the walls of blood vessels allowing finding the optimal extravasation site,⁴⁶ activation of endothelial cells by CTCs to create gaps in the endothelial layer to facilitate their extravasation accomplished through the secretion of chemokines, cytokines,⁴⁷ then diapedesis or transmigration of CTCs through endothelial gaps and final extravasation in the target organ and establishment of

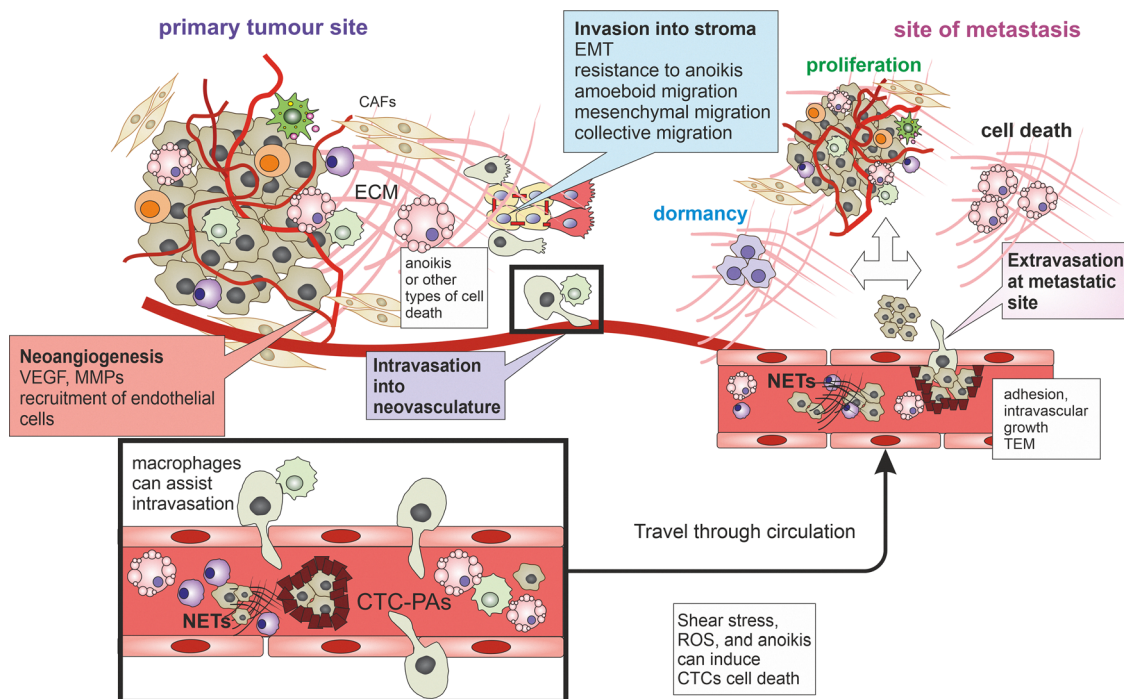


Fig. 1 Scheme of the metastatic process. The sequential stages involved in the tumor metastasis process start at the primary tumor location and lead to the formation of secondary metastatic lesions. At the primary tumor location, cancer cells engage with the ECM and cancer-associated fibroblasts (CAFs) promoting tumor expansion and angiogenesis *via* elements like VEGF and matrix metalloproteinases (MMPs). Tumor cells undergo epithelial-to-mesenchymal transition (EMT) to acquire invasive potential, allowing them to infiltrate the stroma and resist anoikis. In the stroma, cancer cells invade the newly developed blood vessels. Circulating tumor cells (CTCs) face shear stress, reactive oxygen species (ROS), and various obstacles while travelling through the bloodstream, where they can create platelet aggregates (CTC-PAs) or engage with neutrophil extracellular traps (NETs) to improve their survival. At distant sites, CTCs extravasate into the new microenvironment, propelled by adhesion and transendothelial migration (TEM). These spread tumor cells can enter a dormant state, may undergo cell death, or begin to proliferate and form secondary tumors.



a new tumor.⁴⁸ One of the proteins involved in the homing process is metadherin (MTDH). MTDH was shown to promote the attachment of CTCs to the extracellular matrix components of the target site and increase their invasive potential by promoting invadopodia formation.⁴⁹ Furthermore, MTDH can modulate the activity of various key signaling pathways including PI3K/AKT and NF- κ B, further supporting metastatic progression.⁵⁰

TEM, however, is not always present as arrested CTCs have been found to proliferate directly in blood vessel lumina leading to their rupture by large intraluminal cancer colonies.⁵¹ Finally, carcinoma cells that successfully managed to extravasate seem to be sentenced to either their elimination from the parenchyma of the tissue or to enter the state of dormancy in which they remain as single CTCs or micrometastatic clusters for extended periods of time before they advance into detectable metastases. A summary of the metastatic process is shown in Fig. 1.

1.4. Forming a pro-metastatic microenvironment at secondary sites

Previous studies suggest that the vast majority of disseminated tumor cells (DTCs) do not survive circulation and only less than 0.2% of them form metastasis.⁵² The final destination of CTCs is determined by several factors. The main phenomenon describing the preferential migration or colonization is termed organ-specific tropism. Such selectivity is driven by many cellular and molecular mechanisms, including communication between the cancer cells and the secondary tumor site. This communication is governed by specific chemokine signaling pathways, the most well-known being CXCR4 receptors on CTCs and CXCL12 located in the bone marrow, or the CCR5–CCL5 axis facilitating lung metastasis.^{53,54} Other strong indicators are adhesion molecules guiding the whole CTC transport, mainly α 4 β 1 (breast CTC adhesion to the endothelium of the bone marrow) and α 5 β 1 (liver adhesion) integrins⁵⁵ and E-/P-selectins facilitating the rolling of CTCs along the blood vessel walls.⁵⁶ The secondary tumor site is also being primed in advance to ease the extravasation and the formation of metastases. This process of priming on a specific site creates a pre-metastatic niche. Some of the most studied driving forces of this priming are extracellular vesicles (EVs). EVs encapsulating many molecular signals are sent from the primary tumor to alter the secondary microenvironment.⁵⁷ The pro-tumorigenic pre-metastatic niche is characterized by a modulated ECM, collagen crosslinking, local inflammation supporting dual functions (both anti- and pro-tumorigenic) of immune cells, and growth factors facilitating cancer cell survival and growth. Recent studies revealed other factors in this dynamic complex priming including the synergy of B cells and T cells creating an immunosuppressive environment⁵⁸ and tissue remodeling by neutrophils,⁵⁹ highlighting the dual role of immune factors. Potential therapies targeting the pre-metastatic niche and disrupting the primary tumor-secondary site communication have been investigated as an additive novel therapeutic avenue to tumor eradication.⁶⁰

The new tissue microenvironment also presents a challenge since it is devoid of the familiar ECM constituents, growth factors, and stromal cells of the primary site. Poor adaptation often results in dormancy, a prolonged state of growth arrest that reflects a failure of tumor cells to adapt and proliferate in secondary tissues.⁶¹ There are several types of dormant states including cellular dormancy,⁶² angiogenic dormancy,⁶² immune-mediated dormancy,⁶³ microenvironment-mediated dormancy,⁶⁴ or therapy-induced dormancy.⁶⁵ During cellular dormancy, the cell cycle of DTCs is arrested and their metabolic activity is reduced. Angiogenic dormancy occurs before the angiogenic switch, in which DTCs depend only on the pre-existing vasculature and oxygen supply. Immunosurveillance composed mainly of cytotoxic T-cells and natural killer cells can eliminate DTCs and maintain them in an immune-mediated dormancy state. Microenvironment-mediated dormancy depends on microenvironmental signals inhibiting proliferation and promoting dormancy. Such dormancy states can also emerge because of targeted therapy or chemotherapy, however, there is a high risk of developing resistance to treatment and subsequent tumor relapse. The switch from dormancy to active proliferation may be initiated by a range of environmental factors.

2. Intrinsic and extrinsic factors in the metastatic setting

The formation of metastases is a complex process dependent on intrinsic factors inside cancer cells and extrinsic factors of the surrounding tumor microenvironment. The spread of cancer is driven by numerous genetic mutations and alterations in cellular metabolism enabling cancer cells to acquire the capability to colonize distant sites. The total measure of genetic alterations present in tumor DNA is termed tumor mutational burden (TMB) and is often expressed as the number of mutations per megabase (Mb) of DNA.⁶⁶ High TMB indicates a significant accumulation of genomic instabilities, leading to the production of aberrant proteins and at the same time promoting the generation of neoantigens for immune recognition.⁶⁷ Therefore, high TMB is often associated with elevated responsiveness to immunotherapies.⁶⁸ On the other hand, the higher the number of mutations, the higher the tumor cell population diversity, enabling convenient subclones to initiate metastases.⁶⁹ Mutations providing a selective growth advantage to tumor cells directly contributing to the progression of the disease are referred to as driver mutations.⁷⁰ Their identification is crucial for the development of targeted treatments. *KRAS* as well as *TP53* mutations, found in NSCLC, colorectal, and pancreatic cancer, have been linked to increased risk of metastasis.^{71,72} Currently, targeted inhibitors against *EGFR* and *BRAF* mutations have been successfully used in the treatment of metastatic cancers characterized by these genetic alterations.^{73,74} Nevertheless, their sustained efficacy can be limited by the development of resistance. Gene expression patterns in metastatic sites are influenced also by epigenetic



alterations. In this context, potential epigenetic markers have been identified in metastatic breast cancer,⁷⁵ DNA methylation has been linked to treatment resistance in metastatic prostate cancer,⁷⁶ the role of lncRNAs has been discussed in metastatic melanoma,⁷⁷ and chromatin remodeling in metastatic ovarian cancer,⁷⁸ and many more. Given that epigenetic changes are reversible, targeted therapies aimed at modifying or completely reversing these changes are being developed, making them an appealing target in cancer therapy.

The highly dynamic and adaptable nature of cancer cells is facilitated by their exceptional phenotypic plasticity. This feature allows reversible switching between cellular states, contributing to the tumor heterogeneity. Genetic and epigenetic alterations and programs mentioned above define specific cellular states and their biological properties, increasing phenotypic plasticity. These switches, interchangeable over time, lead to the phenotypic versatility of cancer cells promoting metastatic progression and having profound implications for the course of the disease. The most extensively studied example of phenotypic plasticity is the epithelial-mesenchymal/mesenchymal-epithelial (EMT-MET) process. However, hybrid state cancer cells, also referred to as “quasi-mesenchymal cells”, are considered the most aggressive cell phenotype.^{79,80} This cellular shift in the epithelial–mesenchymal spectrum, primarily characterized by the expression of CD51, CD61, and CD106 proteins, allows them to adapt to many different environments, ultimately leading to the evolution of cells with the highest metastatic potential.⁷⁹ Cancer cells undergoing EMT are also less responsive to cancer therapies, underscoring the significant role of phenotypic plasticity in contributing to the development of drug resistance.⁸¹ Furthermore, cancer cells exhibit dynamic transitions between cancer stem cells (CSCs) and differentiated cancer cells.⁸² The interplay of CSCs, shifting between their mesenchymal phenotype with enhanced invasive metastatic properties and their epithelial phenotype, which facilitates the formation of secondary sites, is a critical determinant of colonization.

Throughout the metastatic cascade, cancer cells encounter diverse environments with variations of oxygen availability, nutrient resources, and metabolites, exerting substantial external pressures on their metabolic states. This pressure affects both enzymatic machinery and substrate utilization. The metabolic plasticity during metastatic colonization allows cancer cells to adapt to the microenvironment of the secondary organs and their prevailing nutrient profile. For instance, in a mouse model of breast cancer lung metastasis, there is an abundance of pyruvate in lung interstitial fluid.⁸³ Therefore, pyruvate uptake, mainly fueling pyruvate-dependent anaplerosis, is a crucial factor supporting the successful colonization and outgrowth of lung metastasis. In another study, fatty acid metabolism mediated by fatty acid binding protein 5 (FABP-5), was found to notably support lymph node metastases in a mouse model of cervical cancer.⁸⁴ Overall, these findings collectively illustrate that in each distant metastatic site cancer cells undergoing the metastatic cascade exhibit distinct metabolic phenotypes, underscoring their remarkable metabolic plasticity.

The major components of the TME contributing to tumor metastasis are cancer-associated fibroblasts (CAFs). Their most powerful cancer-promoting mechanisms include ECM remodeling by secretion and modification of collagen, laminin, and fibronectin, the production of MMPs allowing cancer cells to migrate through the ECM, and the production of VEGF, which directly stimulates angiogenesis. Additionally, CAFs secrete many pro-tumorigenic factors, including chemokines that attract immunosuppressive cells. CAFs, therefore, represent a promising target for both anticancer or even antimetastatic therapies due to their critical role in tumor progression and immune modulation. These potential therapies aim to reduce their pro-tumorigenic functions, induce phenotypic switching and lock CAFs in the tumor-restrictive state, blocking their differentiation, or targeting a specific CD10⁺GPR77⁺ CAF-subset and eliminating them by targeted ablation.⁸⁵ Recently, metastasis-associated fibroblasts (MAFs) were identified as another subgroup contributing mainly to cancer metastasis, establishing metastatic niches and mediating resistance to therapy.⁸⁶ Unlike CAFs, MAFs emerge specifically at the metastatic sites, including brain metastases, in which fibroblasts are usually absent under normal physiological conditions.⁸⁷ Another ECM component promoting metastasis is tumor-associated macrophages (TAMs). TAMs are key cell subsets producing chemokines, cytokines and growth factors, contributing to the establishment of an immunosuppressive TME. Moreover, TAMs are abundantly present in EMT hotspots, therefore their activity is directly linked to the EMT process.⁸⁸ They are also critical aspects of the angiogenic switch. It was shown that the absence of TAMs leads to a 40% reduction in blood vessel density.^{89,90} CAFs, MAFs, and TAMs altogether facilitate secondary lesion formation by promoting a pre-metastatic niche. TAMs help recruit CAFs to the secondary site, and both modulate immune responses and provide structural support for metastatic formation.⁹¹ MAFs arise in response to communication between fibroblasts and macrophages.⁹² Altogether they provide complex machinery to ease extravasation and formation of metastasis. The main targetable intrinsic and extrinsic factors involved in the metastatic setting are shown in Fig. 2.

The migration of cancer cells is ensured by substantial cytoskeletal reorganization.⁹³ The cytoskeleton responds to external signals orchestrating complex behavior such as the formation of lamellipodia, pseudopodia, and, principally, the process of migration. The migration modes also depend on the stiffness and composition of the extracellular matrix.⁹⁴ The motility of cancer cells is mainly enabled by the rearrangement of actin and intermediate filaments, microtubule dynamics, and focal adhesions. Contractile forces generated by actin and myosin interactions aid tumor cells to squeeze through tight gaps in the ECM.⁹⁵ These forces are particularly challenging for the nucleus of the cell, which is the stiffest organelle determining the extent of possible compression of the whole cell. However, this extensive compression of the cell and the nucleus threatens genome stability and often leads to nuclear rupture. This rupture can be prevented by



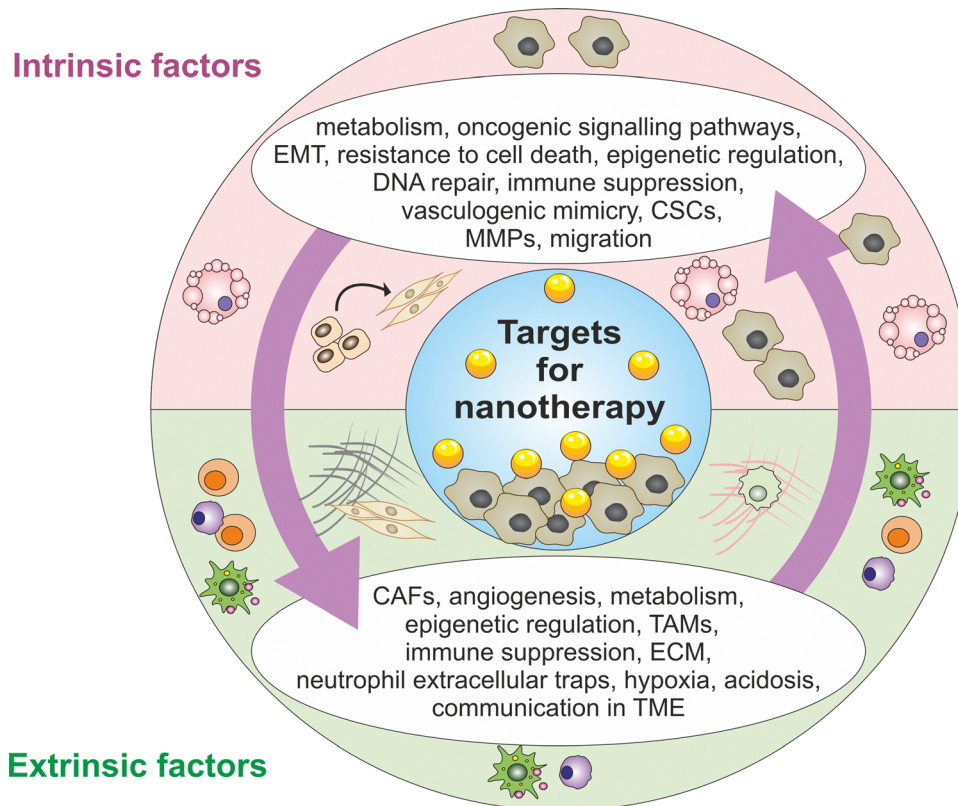


Fig. 2 Overview of targetable intrinsic and extrinsic factors in the metastatic setting. The interplay between intrinsic molecular alterations and external influences of the tumor microenvironment defines the metastatic potential of cancer cells. Abbreviations: EMT – epithelial–mesenchymal transition, CSCs – cancer stem cells, MMPs – matrix metalloproteinases, CAFs – cancer-associated fibroblasts, TAMs – tumor-associated macrophages, ECM – extracellular matrix, TME – tumor microenvironment.

nucleus-centrosome linkage, which senses the tension and facilitates endosomal vesicles trafficking to the cell leading edge, delivering membrane type 1 matrix metalloprotease (MT1–MMP).⁹⁶ However, if the nucleus indeed breaks open, this rupture causes severe DNA damage and genomic instability which, may further promote cancer progression.⁹⁷ For example, it was shown that DNA-destroying enzyme three prime repair exonuclease 1 (TREX1) vastly adds to the invasiveness of the cancer cell.⁹⁸ On the other hand, this phenomenon may lead to the development of novel antimetastatic drugs by specifically targeting these cancer cells and preventing them from recovery. Microtubules establish cell polarity, an essential factor in guiding controlled and directed cancer cell invasion.⁹⁹ During EMT, the expression of vimentin, as an intermediate filament protein, is upregulated in tumor cells, providing mesenchymal cells with structural support, ensuring anchorage of organelles during migration,¹⁰⁰ and facilitating cell adhesion to a substrate.¹⁰¹ Focal adhesions, which serve as the cell anchor points, are formed by cell surface integrins binding to ECM components and enabling cells to apply force and relocate.¹⁰² Therapeutic strategies aimed at disruption of cytoskeletal dynamics are actively under investigation as potential therapeutic approaches limiting the progression of metastases.^{103,104}

3. Antimetastatic treatments

Targeting metastatic cancer involves a broad spectrum of approaches interfering with the ongoing metastatic formation. Micrometastases refer to small clusters of cancer cells in secondary sites that are hardly detectable due to their size, therefore, micrometastases are often only assumed to be present and their treatment is mostly preventive. In contrast, macrometastases are detectable through physical examination or imaging and are the main focus of treatment efforts. Predominant systemic approaches for both micro- as well as macro-metastasis are chemotherapy, immunotherapy, and targeted therapy or combinations thereof. Eliminating metastasis in the vicinity of primary tumors can be achieved *via* radiation, whereas distant metastases are generally managed by chemotherapy. In some cases, such as metastatic breast cancer afflicted axillary lymph nodes, surgical resection is also an option.¹⁰⁵ Chemotherapy remains the standard of care across a broad spectrum of malignant tumors. However, recent studies suggest that chemotherapy can even promote metastasis through multiple mechanisms, such as promoting cells with highly metastatic phenotypes, activation of EMT, or induction of cancer stem cell populations.^{106,107} Moreover, chemotherapeutics greatly affect the hematological system as well as



immune responses, inflammatory responses, and overall ECM of tumor tissue.^{108,109} For example, paclitaxel increases invasive behavior by inducing invadopodia and at the same time facilitates vascular alterations such as blood vessel leakage or overall vascular permeability leading to angiogenesis.¹¹⁰ Current cytotoxic chemotherapies lack complementary approaches targeting the metastatic cascade. Therefore, in metastatic settings, combination therapies of broad-spectrum cytotoxic chemotherapies together with antimetastatic treatments are crucial for higher survival rates and for overall reduction of metastatic burden.

The early therapeutic target in the TME was tumor neovasculation. The clinically approved angiogenesis inhibitor bevacizumab has shown efficiency in metastatic settings by binding vascular endothelial growth factor (VEGF).¹¹¹ This leads to the inhibition of angiogenesis and overall restriction of tumor growth by depriving it of the blood supply. Moreover, the addition of bevacizumab to combination fluorouracil-based chemotherapy, termed FOLFOX, significantly improved the survival of metastatic colorectal cancer patients.¹¹² Matrix metalloproteinase (MMP) inhibitors emerged as another promising therapeutic target of metastatic cancers since dysregulation and overexpression of MMPs are associated with aggressive disease and poor outcomes.^{113,114} Unfortunately, to this day, no MMP inhibitor has been approved for clinical use due to their severe disruption of healthy tissue homeostasis.^{115,116} Clinical failure of MMP inhibitors may also be related to the ability of tumor cells to switch between MMP-dependent mesenchymal movement, and MMP-independent amoeboid motility. This switch provides cancer cells with an alternative invasion pathway in cases when MMP activity is inhibited.¹¹⁷ Overexpressed C-X-C chemokine receptor type 4 (CXCR-4) is also highly involved in cell migration and metastasis.¹¹⁸ The interaction of CXCR-4 and its ligand CXCL-12 plays a pivotal role in the recruitment of tumor cells in CXCL-12-rich distant sites such as lymph nodes or bone marrow.¹¹⁹ CXCR-4 inhibitors block the CXCR-4/CXCL-12 axis and inhibit metastatic formation. Many preclinical investigations and clinical trials in this context have investigated the CXCR4 antagonist AMD3100 (Plerixafor) in the context of metastatic breast cancer,¹²⁰ metastatic colon cancer,¹²¹ and hematological cancers.¹²² Other potential CXCR4 inhibitors, such as BL-8040 (Motixafortide), have been evaluated for non-small lung cancer¹²³ and pancreatic cancer metastasis.¹²⁴ A clinically approved class of antimetastatic drugs are tyrosine kinase inhibitors (TKIs). TKIs interfere with metastasis driving signaling pathways, thereby halting or slowing down the disease progression. TKIs specifically target abnormal signaling pathways that drive cancer progression by inhibiting tyrosine kinase phosphorylating tyrosine residues. This disruption prevents downstream signaling, thereby inhibiting tumor expansion and metastasis.¹²⁵ TKIs clinically approved for the treatment of metastatic cancers are, for example, sunitinib (brand name Sutent)¹²⁶ for the treatment of metastatic renal cell carcinoma, lapatinib (brand name Tyverb) for metastatic HER2-positive breast cancer,¹²⁷ and regorafenib (brand name

Stivarga) for metastatic colorectal cancer.¹²⁸ These treatments are often used in combination with chemotherapy, radiation therapy, or immunotherapy.

Historically, cancer treatment has primarily focused on the intrinsic properties of tumor cells. However, the advent of immunotherapy has underlined the undeniable significance of the tumor microenvironment in the treatment of metastasis. Immune checkpoint inhibitors (ICIs) revolutionized the treatment of metastases by their ability to block proteins inhibiting the recognition of cancer cells by the immune system and their subsequent attack.¹²⁹ The most well-known checkpoint proteins muting the immune system are programmed cell death protein (PD-1) and cytotoxic T-lymphocyte-associated protein 4 (CTLA-4).¹³⁰ Metastatic cancers that respond to ICIs, such as metastatic melanoma, usually have a high TMB. These tumors generate many mutated peptides displayed on the tumor's major histocompatibility complex (MHC) and are identified by the immune system as foreign "neoantigens".¹³¹ Another therapeutic strategy demonstrating promising results especially in hematological malignancies is chimeric antigen receptor T-cell therapy (CAR-T). In this process, the autologous T cells are reprogrammed to target antigens specific for cancer cells. In B-cell malignancies, the T cells used for CAR-T are mostly designed to target CD19.¹³² Another immunotherapy involving patient-derived T cells is tumor-infiltrating lymphocyte (TIL) therapy, in which autologous T cells isolated from the patient's tumor are expanded *ex vivo* in the lab and then reinfused into the patient. TIL therapy resulted in long-term remission of patients with metastatic melanoma.^{133,134}

The novel term "migrastatics" has been introduced for drugs interfering with the ability of cells to metastasize.^{135,136} Among them, pentamethinium salts, initially designed as mitochondrial probes, and curcumin derivatives, derived from turmeric, emerged as very promising therapeutics in preclinical research. Pentamethinium salts demonstrated a significant impact on mitochondria, effectively inhibiting cancer cell proliferation and migration *in vitro*. This mechanism is likely attributed to the inhibition of dihydroorotate dehydrogenase (DHODH)-dependent respiration. Moreover, pentamethinium salts have been shown to induce oxidative stress, alter mitochondrial distribution, and decrease mitochondria mass.¹³⁷ Curcuminoids or curcumin derivatives are also associated with the inhibition of cancer cell migration and invasion. Curcumin has been identified as an inhibitor of NF- κ B signaling as well as many other signaling pathways linked to cancer cell motility.¹³⁸ Additionally, curcumin reduces levels of hypoxia-induced factor 1 (HIF-1) subsequently reducing VEGF production.¹³⁹ Curcumin also downregulates the antiapoptotic protein Bcl-xL, and therefore enhances cancer cell susceptibility to apoptosis.¹⁴⁰ The combined anticarcinogenic, anti-inflammatory, and anti-oxidative effects of curcumin and its derivatives highlight their potential as promising anticancer therapeutics. Another group of potential migrastatics are multikinase inhibitors designed to inhibit Rho-associated protein kinase (ROCK) and myotonic dystrophy kinase-related Cdc42-binding kinase (MRCK), both involved in cytoskeletal dynamics and the motility of cancer



cells.¹⁴¹ In general, combining migrastatics targeting the invasiveness of the cells with antiproliferative therapies could result in a highly effective synergic approach for treating metastatic cancers. Optimal migrastatic targets are likely those essential to all cell migration forms, such as cytoskeletal reorganization, ATP production, mitochondrial metabolism, and cellular contractility.¹⁴² Moreover, halting metastatic dissemination by migrastatics can also decrease the need for cytotoxic therapies in high doses.

Nowadays, many investigational approaches aim to develop advanced combination therapies. By simultaneously targeting two distinct cellular targets, these bispecific therapies aim to leverage dual-target recognition to achieve synergistic effects, enhanced selectivity, and reduced off-target toxicity compared to monospecific approaches. This is achieved for example by using bispecific antibodies simultaneously targeting cancer cell antigens and an immune cell receptor, or by employing nanoparticle delivery systems designed to deliver agents to the TME.

4. Nanotechnology-based approaches for metastatic cancer treatment

The application of nanotechnology in medicine, known as nanomedicine, has emerged as a breakthrough approach for more precise and early disease diagnosis, including cancer, while also serving as a powerful tool for effective therapy. To this day, several nano-based carriers have been described in the medical field, including liposomes, polymeric micelles, quantum dots, golden nanoshells, and many more. Moreover, the coupling of nano-drugs with targeted nanoscale delivery systems has led to the development of advanced nanoscale devices – nanorobots – engineered to perform precise therapeutic actions. Considering their tunable physical and chemical characteristics, which allow for a wide spectrum of therapeutic functions, these nanomachines can serve as both detection agents for specific biomarkers or disease locations and targeting agents delivering therapies to targeted sites. Furthermore, nanotechnology revolutionized personalized medicine in terms of reducing severe side effects of conventionally used chemotherapy. This was achieved through the modulation of biodistribution, thereby improving the selective delivery and overall balance between drug efficiency and toxicity. These advances may enable the overcoming of drug resistance and improvements in biocompatibility. For instance, golden nanoparticles embedded in the hydrogel silencing the multidrug-resistance-associated protein 1 (MRP1) gene were reported to prevent the efflux of the chemotherapeutic 5-fluorouracil and thereby overcome 5-FU treatment resistance.¹⁴³ Additionally, nanotherapeutics have the potential to overcome biological barriers, such as the blood–brain barrier (BBB), dense extracellular matrix, and high tumor interstitial fluid pressure. In recent years, nanotechnology has reintroduced the concept of theranostics, which combines diagnostic procedures with therapeutic interventions.

The term “nanotechnology” inherently refers to the most significant feature of such a delivery system, its nanoscale size. Nanoparticles (NPs) typically range from 1 to 100 nm in size. The efficiency of NP delivery can be adapted by modulation of the size and surface characteristics of NPs. However, a significant drawback of nanocarriers is their early removal from circulation through reticuloendothelial system (RES) systems. Nevertheless, this limitation may be resolved using various strategies *e.g.*, by employing highly hydrophilic polymeric materials.¹⁴⁴ After being altered by hydrophilic polymers, NPs can remain in circulation for a prolonged time which increases the possibility for their interaction with cancer cells.¹⁴⁵

Anticancer drugs are mostly delivered to tumor sites by injection into the tumor parenchyma or systemic circulation. Administration of anticancer drugs into systemic circulation is considered easier and more acceptable to patients. However, since targeting specific areas throughout the distribution by systemic circulation remains challenging and possible side effects often occur, this technique is not always effective. Moreover, medications often accumulate at the tumor periphery in low concentrations, failing to effectively penetrate and eradicate the tumor. This can result in tumor recurrence or metastasis.¹⁴⁶ Furthermore, while being transported, anticancer drugs may be metabolized, bind non-specifically to proteins or other tissue components. The strategies used to enhance the specificity of nanoparticles (NPs) for targeting tumor sites include both active and passive targeting mechanisms. Both are essential for increasing the potency of treatments while reducing the adverse effects. For passive targeting, the key aspect is termed the enhanced permeability and retention (EPR) effect.¹⁴⁷ The EPR effect depends on specific features of tumor vasculature, specifically, its leaky and irregular nature. This tissue characteristic allows NPs to passively accumulate in tumor tissue. The ideal size range of NPs for effective passive targeting and leaking out of the vasculature is in the range of 10 to 200 nm. Also, the extent of NP accumulation at the tumor site is increased with their prolonged circulation in the bloodstream. Nevertheless, the EPR effect is variable, as it is influenced by heterogeneity among tumor types. Small metastases, with sizes below 100 mm³, with poor vascularization cannot be accessed by EPR.¹⁴⁸ By passive tumor targeting the NPs are unable to directly target malignant cells.¹⁴⁹ Several clinically approved nanoformulations of traditional anticancer drugs rely on passive targeting, such as daunorubicin (DaunoXome[®])¹⁵⁰ or paclitaxel (PTX; Genexol PM[®], Abraxane[®]).¹⁵¹ On the other hand, active targeting depends on the functionalization of nanoparticle surfaces with specific antibodies or ligands. These components interact with cancer cell surface receptors and after the NP binding, the therapeutic cargo is released into the cell. These specifically designed ligands therefore improve the targeting specificity of NPs which allows for the generation of novel innovative therapeutic and diagnostic agents. Since endocytosis absorbs NPs selectively after their attachment, selective binding improves the drug penetration while attenuating off-target toxicity.¹⁵² However, as stated before, the efficacy of active targeting can also vary. This variability can be



attributed to the tumor heterogeneity and variability of receptor expression. A specific example of modified nano-delivery systems that utilize active targeting principles is those modified with arginine–glycine–aspartic acid (RGD) peptides.¹⁵³ RGD motifs recognize integrins.¹⁵⁴ These are overexpressed on tumor cells and are highly involved in the process of cancer invasion and metastasis.¹⁵⁵ RGD-recognized integrins are the most common integrins in terms of cell adherence to the ECM. The expression of RGD-binding integrins, specifically $\alpha_5\beta_1$ and α_v , in tumor vasculature inspired the initial attempts to develop specific agents using RGD for reducing tumor angiogenesis and metastasis.¹⁵⁶ Unfortunately, despite promising preclinical results, these findings have not yet been consistently replicated in clinical trials. Intense research continues to explore the potential of RGD ligands to inhibit adhesion, migration, and survival of cancer cells. Another receptor widely overexpressed on cancer cell membranes is the transferrin receptor. Therefore, NPs functionalized with its ligands preferentially accumulate in transferrin receptor-overexpressing tumor cells.¹⁵⁷ Other customized NP delivery systems target glycoproteins, folate receptors, or EGFR.¹⁵⁸ The multistructural glycoprotein CD44 found on the cell surface is primarily involved in inflammation, cellular motility, and cell-to-cell interactions.¹⁵⁹ The active targeting of the CD44 receptor using nanocarriers has therefore been widely studied.¹⁶⁰ CD44 receptors have shown selective binding to hyaluronic acid and chondroitin sulphate functioning as a substrate. Once the CD44 receptor binds to hyaluronic acid,^{161,162} it is activated and leads to increased tumor growth. An innovative NP platform for the treatment of highly invasive triple-negative breast cancer (TNBC) overexpressing CD44 integrates CD44-mediated therapy combined with JAK/STAT inhibitors synergistically disrupting CD44 signaling and blocking JAK/STAT-mediated pro-tumor inflammation, both key drivers of cancer resistance. Polymeric NPs loaded with CD44-targeted NPs significantly reduced cell viability with association to a high dose reduction index (DRI), quantifying the fold reduction in drug dose required to achieve the same efficacy when used synergistically.¹⁶³

To date, NPs of various nature have shown promising results in both metastatic cancer as well as carcinoma *in situ*. The first nanotechnology-based drugs that gained FDA approval were Doxil (1995),¹⁶⁴ a pegylated liposomal doxorubicin, and Abraxane (2005), a protein-bound paclitaxel formulation.¹⁶⁵ In a recent article, it was further reported that drug-free polystyrene NPs alone attach tightly to two different cell-derived structures associated with migration: retraction fibers (RFs) and migrasomes.¹⁶⁶ These structures are typically located in the posterior region of tumor cells during migration. Cell adhesion, motion range restriction, and morphology seem to be affected by this interaction. Through mechanical interaction with the lipid raft/caveolae substructures, NPs coat and form a hard shell on the surface of migrasomes and retraction fibers. Consequently, NPs prevent migrasomes from being recognized, endocytosed, and removed by the surrounding cancer cells or other cells within the TME. As a result, NPs disrupt the role of migrasomes in the process of cancer cell migration.

In recent years, many studies have been conducted on the improvement of efficacy and delivery of many conventionally used therapeutics targeting various aspects of metastatic pathways by addressing their poor water solubility or low bioavailability. Synergistic therapy of NPs containing sorafenib and ursolic acid has been developed for potential advanced treatment of hepatocellular carcinoma.¹⁶⁷ Since sorafenib is poorly soluble in water and is extensively metabolized by CYP3A4 its bioavailability is very low. To improve the bioavailability of sorafenib, another study generated sorafenib polymeric NPs exhibiting a significant impact on lung and breast cancer *CDKN1A* and *STMN1* gene expression, as well as an increase in DNA damage in model cancer cell lines.¹⁶⁸ The delivery of another previously mentioned drug bevacizumab was enhanced by PEGylated cationic liposomes, PEGylated NPs,¹⁶⁹ as well as poly(D,L-lactic-co-glycolic acid-PLGA) NPs. In an *in vivo* glioblastoma model, these PLGA NPs demonstrated higher anticancer activity and tumor reduction in comparison to free bevacizumab.¹⁷⁰ Similarly, sunitinib malate activity was boosted by lipid polymer hybrid NPs. In an *in vitro* breast cancer model, sunitinib loaded NPs exhibited significant cytotoxic activity, as evidenced by increased p53 and caspase-3/9 activities.¹⁷¹ Lapatinib, another tyrosine kinase inhibitor, exhibited enhanced effectivity in breast cancer when encapsulated into PEGylated lipid layer NPs containing a polymeric core. This nanoparticle design allowed for enhanced cellular internalization and effective induction of apoptosis.¹⁷² In another study, lapatinib/pseudolaric acid B@Ferritin NPs were designed for the therapy of triple-negative breast cancer sensitive to ferroptosis. In both *in vitro* and *in vivo*, these structures showed a more potent tumor suppression accompanied by the lipid peroxidation.¹⁷³

Immune checkpoint inhibitors are also being conjugated to several nanostructures. For instance, a CXCR4 inhibitor Plerixafor together with PD-L1 inhibitor BMS-1 were co-loaded on Cu_2MoS_4 (CMS)/PEG NPs for combined immunotherapy of pancreatic ductal adenocarcinoma. This combination enhanced CXCR and PD-L1 inhibition relative to the free inhibitors while remodeling the TME by increasing tumor infiltration of $\text{CD4}^+/\text{CD8}^+$ T cells, and at the same time reducing immunosuppressive cells.¹⁷⁴

Latest advances in cancer nanotechnology have led to the categorization of its applications into several focused strategies designed to reduce metastatic spreading. These include targeting the process of tumor cell invasion and intravasation, targeting dissemination and migration, targeting micrometastasis and pre-metastatic niche, and targeting macrometastasis.

Targeting the process of tumor intravasation is widely investigated as a metastatic treatment strategy. For example, nano-formulated quinacrine (NQC), originally an anti-malarial drug, combined with ADAM-17 inhibitor (GW280264) was shown to decrease the invasion rates in cervical cancer stem cells. This combination therapy targets the increased expression of Nectin-4, a protein linked to the Notch signaling pathway and possibly responsible for 5-FU resistance in metastatic models, by blocking its nuclear translocation.¹⁷⁵ Furthermore,



the co-prodrug-loaded micelles (CLMs) containing cathepsin B/pH dual-sensitive block copolymer conjugated with doxorubicin and loaded with nifuroxazide were synthesized and explored as potential breast cancer metastasis nanomedicine. CLMs were shown to inhibit migration and invasion *in vitro* and *in vivo* orthotopic and lung metastasis breast cancer mice models demonstrating significant antimetastatic effects.¹⁷⁶ In another study, an intravenous folate-conjugated nanocarrier loaded with miR-125b-5p plasmid effectively inhibited the invasiveness of hepatocellular carcinoma *via* the Wnt/ β -Catenin pathway.¹⁷⁷

Migration disruption could be another approach to halting the metastatic cascade. Di-2-pyridylketone-4,4-dimethyl-3-thiosemicarbazone (Dp44mT) and cisplatin-co-loaded NPs have been shown to suppress VEGF, HIF-1, vimentin, and MMP2 expression. Dp44mT is an iron chelator inhibiting cell migration. In an *in vivo* model, this nanoplatform significantly inhibited lung metastasis as well as overall tumor growth.¹⁷⁸ This nanoformulation is another example of combining conventional functional drugs together with an antimetastatic approach encapsulated in a nanocarrier for enhanced tumor intracellular drug accumulation and therefore, reduction of side effects. In another example, etoposide, a topoisomerase II inhibitor, was combined with a layered double hydroxide (LDH) carrier, which significantly inhibited cancer migration and invasion *in vitro*.¹⁷⁹

The strategy of targeting the pre-metastatic niche was also demonstrated as promising. As mentioned earlier, exosomes play a crucial role in establishing a pre-metastatic niche. CBSA/siS100A4@Exosome nanoparticles (cationic bovine serum albumin conjugated siS100A4 and exosome membrane coated nanoparticles) exhibited potent gene-silencing effects inhibiting malignant breast cancer cells and demonstrating strong affinity for lung metastasis.¹⁸⁰

Macrometastases remain the primary focus of the majority of nano-antimetastatic studies. In a recent study, NPs loaded with docetaxel and sonic hedgehog protein (SHH) siRNA were used for bone metastasis targeting in prostate cancer. After reaching the bone microenvironment, they inhibited cancer cell proliferation and migration by SHH autocrine/paracrine inhibition, increasing the anticancer effects of docetaxel.¹⁸¹ A bone-targeted nanodelivery system was also demonstrated in breast cancer bone metastasis. Gold nanorods inside silica nanoparticles combined with zoledronic acid showed specific bone-targeting *in vivo* as well as reducing VEGF levels and inducing apoptosis of cancer cells.¹⁸² The previously mentioned AMD3100, a CXCR4 antagonist, was also conjugated with NPs and *proMel*, the artificial gene encoding prodrug form of melittin releasing cytolytic melittin once activated by MMP-2 present in tumors. In a breast cancer brain metastases mouse model, poly(lactone-co- β -amino ester)-AMID3100-*proMel* nanoparticles effectively inhibited tumor progression, establishing novel promelittin-mediated gene therapy.¹⁸³

Nanotherapeutics can also target components of the TME. Several NP platforms are actively targeting MAFs. In liver cancer, the equivalent to CAFs or MAFs are hepatic stellate

cells (HSC)/myofibroblasts presenting a great target for potential stroma-based therapies. Surface-modified nanocarriers with cyclic peptide binding to the platelet-derived growth factor receptor beta (PDGFR β) or with mannose-6-phosphate binding to the insulin-like growth factor receptor II (IGFRII) successfully directed NPs loaded with drugs to HSC/CAF *in vivo*.¹⁸⁴

In clinical practice, a variety of tumor ablation techniques are currently used to treat cancer, including thermal, cryo-, microwave, and ultrasound ablation. These techniques are valued for their reduced trauma to the affected area, site-specific tumor cell killing, and controllability. Apart from the immediate lethal impact, the eventual death of tumor cells following tumor ablation serves as a possible source of antigens to initiate an immune reaction.¹⁸⁵ Both primary and metastatic cancers can be treated with thermal ablation by inducing an immune response.¹⁸⁶ This concept is also relevant and has been adapted to the nanotechnology-based approaches. Specifically, nanoparticle-based photothermal therapy (PTT) can trigger specific immune responses, such as immunogenic cell death (ICD), release of damage-associated molecular patterns (DAMPs), activation of antigen-presenting cells (APCs), release of cytokines or stimulation of tumor-infiltrating lymphocytes (TILs).¹⁸⁷ However, several recent studies have shown that PTT alone may not be sufficient to limit the growth of primary tumors or distant metastases.¹⁸⁸ The symbiotic interaction between tumor cells and the host immune system introduces additional complexity to cancer immunotherapy. Tumor cells may exhibit tolerance to immune-mediated elimination, or in some cases, even exploit the immune system components to promote their progression. In addition to cell apoptosis and necrosis induced by heat exhaustion, PTT may have additional anticancer effects. PTT, for instance, showed exceptional superiority in overcoming drug resistance. It can eradicate both drug-sensitive and multidrug-resistant (MDR) cancer cells and has shown no discernible signs of resistance in preclinical tumor treatment models.¹⁸⁹ It is well accepted that the overexpression of drug-efflux transporters, *p*-glycoprotein (*p*-gp), and MRP1 contributes to the MDR of various cancers.¹⁹⁰ According to a recent study, drug resistance in A549R cells, a drug-resistant variant of the A549 cell line, may be reversed by inhibiting MRP1 expression using cyanine dye-loaded nanoparticle-based PTT.¹⁹¹ According to another study, PTT based on carbon and gold nanoparticles can help overcome doxorubicin (DOX) resistance by promoting the activation of heat-shock factor trimers, which in turn downregulate *p*-gp.¹⁹² Additionally, PTT may enhance the chemotherapy effectiveness by rupturing the integrity of the cell membrane, leading to higher drug accumulation within the tumor.¹⁹³ Recently, scientists have created a novel active-targeted drug delivery nanoplatform based on copper sulfide nanoparticles (CuS NPs) functionalized with RGD ligand. This system has been designed for laser irradiation capable of deep tissue penetration. It provides an alternating therapy approach combining chemotherapy and PTT resulting in a synergistic 3.53-fold increase in therapeutic effect *in vivo*.¹⁹⁴ This approach also addressed the tumor metastasis



spread caused by hypoxia. The nanoplatform demonstrated high efficiency photothermal conversion, deep tissue heat penetration, and DOX release from nanostructures during laser irradiation. This can be attributed to the simple structure of the platform and the ability to target cells. Additionally, the alternating chemo/PTT technique may prevent tumor liver metastasis by reducing tumor hypoxia, indicating that this nanoplatform may enhance cancer treatment and improve prognosis in preclinical models.¹⁹⁵

Another approach, photodynamic therapy (PDT), has also been studied in the context of nanotechnology and treatment of metastatic cancer. The treatment was demonstrated in a recent study focusing on uveal melanoma (UM), a rare ocular malignancy. Here, ROCK inhibitor was used in combination with PDT based on Ce6-embedded nanophotosensitizers (FIC-PDT) and was further combined with PD-1/PDL-1 blockage. FICs were utilized to boost the effectiveness of PDT, and their application has been explored for their ability to induce ICD. One of the main challenges in UM therapy is metastasis, particularly spreading to the liver, which can be potentially inhibited by a mechanism known as immunogenic clearance—a process in which signals released by dying tumor cells activate a broader immune response to eliminate tumor cells. This combination therapeutic approach stimulated the immunological response and led to the trafficking of CD8⁺ T cells into the initial tumor site. Consequently, anti-PD-L1 antibody plus immunogenic clearance brought on by FIC-PDT and ROCK inhibitor could be a powerful immunotherapeutic approach for UM.¹⁹⁶

Among various nanotechnology applications in the treatment of cancer, NPs coupled with RNA molecules have the potential to greatly upregulate the expression of target tumor suppressor genes or to suppress the expression of target oncogenes. RNA-based tumor therapy is known for its low toxicity and for a lack of contribution to the drug resistance of tumors. However, due to RNases and exonucleases present in the human immune system, such therapeutic molecules degrade rapidly. Therefore, RNA encapsulation in NPs (lipid, inorganic, polymer, or biomimetic NPs) significantly improves cancer therapy effectiveness, particularly for metastatic cancer, where targeted delivery is crucial for reaching distant sites and overcoming treatment resistance. For instance, in melanoma patients, RNA lipoplexes (DNA-carrying nonviral cationic liposomes) comprising 1,2-dioleoyl-*sn*-glycerol-3-phosphatidylethanolamine (DOPE) and *N*-[1-(2,3-dioleoyloxy)propyl]-*N,N,N*-trimethylammonium chloride (DOTMA) lipid carriers were shown to induce T-cell immune responses, and interestingly, mediate powerful interferon α -dependent metastatic tumor rejection.¹⁹⁷ In another example, the overexpression of TWIST-related protein 1 in metastatic carcinomas was successfully suppressed using mesoporous silica nanoparticles (MSNs) modified with hyaluronic acid (HA). HA-modified MSNs designed to deliver siRNA-419 (siTWIST) target CD44 overexpressed on cancer cells. Combined treatment of siTWIST-HA and cisplatin led to noticeably smaller tumors and fewer metastases in mice.¹⁹⁸

Another contribution to anticancer nanotechnology has been triggered by quantum dots (QDs). Due to their broad biocompatibility, high water solubility, and tunable surface functionalities, quantum dots have emerged as a promising platform for medical applications. A recent study has shown that downregulation of matrix metalloproteinase-2 and -9 (MMP-2/9) and vascular endothelial growth factor receptor 2 (VEGFR2) expression *in vitro* by carbon-based quantum dots (CQDs)/Cu₂O can reduce migration and angiogenesis and thereby inhibit tumor progression.¹⁹⁹ The particle internalization significantly reduced the growth and invasiveness of B16F10 melanoma cells. Furthermore, B16F10 cells labeled with quantum dots-GSH can detect the early distribution and migration of B16F10 cells *in vivo* in C57BL/6 mice.²⁰⁰ In another study, tangeretin-zinc oxide (Tan-ZnO) quantum dots, a type of ultra-small quantum dots with dimensions ranging from 5 to 4 nm, were shown to significantly boost cytotoxicity and morphological damage in H358 metastatic human lung carcinoma cells. When compared to the untreated control, the treatment of H358 cells with Tan-ZnO QDs resulted in reduced cell proliferation, cell cycle arrest in the G2/M phase cells, and increased nuclear fragmentation and apoptosis, while significantly inhibiting migration and invasion, both crucial components of metastasis.²⁰¹

Ongoing nanomedicine research has great promise for developing highly specific antimetastatic treatments with many of therapeutical approaches targeting metastases advancing to clinical translation. Inhibitors of crucial migrastatic pathways such as Rho/ROCK, CXCR4, integrins as well as RNA-based approaches aimed at EMT drivers are now nearing clinical applications. As we continue to unravel the complexity of the tumor microenvironment and metastatic pathways, the potential for nanotherapeutics to redefine the treatment of metastatic cancer is becoming increasingly evident. However, further advancements in nanomedicines remain necessary to fully utilize its clinical potential (Tables 1 and 2).

5. Drug uptake

Improving the therapeutic efficacy of nanodrugs remains a pressing issue. Overcoming several biological barriers along the route to the tumor site, as well as achieving efficient extravasation and infiltration in the tumor itself, are major unresolved obstacles. Only a small fraction of injected nanoparticles (often less than 1%) actually reaches solid tumors, calling into question the traditional reliance on the EPR effect. The EPR effect is limited because most human tumors lack the vascular leakiness and tissue conditions necessary for passive nanoparticle accumulation.²¹⁹ The acronym “CAPIR” cascade summarizes the main sequential barriers NPs have to cross along the way into the tumor: circulation, accumulation, penetration, internalization, and release.

The most common route for nanotherapeutics to reach the tumor site is through endocytosis, yet it presents a significant bottleneck for delivery efficiency. Cellular internalization



Table 1 Overview of nanotherapies

Drug name	Nanocarrier	Cargo	Cancer type	Development stage	Ref.
Doxil/Caelyx	PEGylated liposomes	Doxorubicin	Metastatic breast cancer	FDA- and EMA-approved (for Europe as Caelyx)	164
Genexol PM [®] Abraxane [®]	Polymeric micelles Albumin-bound NPs	Paclitaxel Paclitaxel	Metastatic breast cancer Metastatic breast cancer, metastatic pancreatic cancer	Approved in South Korea FDA- and EMA-approved	202 165
CD44 targeted polymeric nanoparticles	Polymeric NPs	Momelotinib + CFM.4.16	TNBC	<i>In vitro/in vivo</i> (preclinical)	163
US NPs	—	Sorafenib + ursulonic acid	Hepatocellular carcinoma	<i>In vitro/in vivo</i> (preclinical)	167
Sorafenib NPs (SFB-PNs)	Polymeric NPs	Sorafenib	Lung cancer, breast cancer	<i>In vitro/in vivo</i> (preclinical)	168
PLGA NPs	PLGA NPs	Bevacizumab	Glioblastoma	<i>In vitro/in vivo</i> (preclinical)	170
Lipid polymer hybrid NPs (LPHNPs)	Chitosan-based lipid-polymer hybrid NPs	Sunitinib malate	Breast cancer	<i>In vitro/in vivo</i> (preclinical)	171
PEGylated lipid layer NPs containing a polymeric core	Core-shell NPs (PEGylated lipid layer + polymeric core)	Lapatinib	Breast cancer	<i>In vitro/in vivo</i> (preclinical)	172
Lapatinib/pseudolaric acid B@Ferritin NPs	Protein-based NPs (ferritin nanocage)	Lapatinib + pseudolaric acid	TNBC	<i>In vitro</i> (preclinical)	173
Cu ₂ MoS ₄ (CMS)/PEG NPs	Hollow mesoporous NPs (PEGylated)	BMS-1 + Plerixafor	Pancreatic ductal adenocarcinoma	<i>In vitro/in vivo</i> (preclinical)	174
Nano-formulated quinaquine (NQC)	PLGA based NPs	Quinaquine	Cervical cancer	<i>In vitro</i> (preclinical)	175
CLMs containing cathepsin B/pH dual-sensitive	Polymeric micelles	Doxorubicin + nifuroxazide	Metastatic breast cancer	<i>In vitro/in vivo</i> (preclinical)	176
Folate-conjugated nanocarrier loaded with miR-125b-5p plasmid	Fa-PEG-g-PEI-SPION (FaPPS)	miR-125b-5b	Hepatocellular carcinoma	<i>In vitro/in vivo</i> (preclinical)	177
Dp44mT (di-2-pyridylketone-4,4-dimethyl-3-thiosemicarbazone) and cisplatin-co-loaded NPs	Polymeric NPs (PEG-PLGA)	Dp44mT + platinum(IV) pro-drug (cisplatin)	Metastatic breast cancer	<i>In vitro/in vivo</i> (preclinical)	178
VP16-LDH	Inorganic layered double hydroxide (LDH) NPs	Etoposide	Lung cancer	<i>In vitro/in vivo</i> (preclinical)	179
CBSA/siS100A4@Exosome NPs	Exosome-based NPs	siS100A4 siRNA	Metastatic breast cancer	<i>In vitro/in vivo</i> (preclinical)	180
NPs loaded with docetaxel and sonic hedgehog protein (Shh) siRNA	Lipid-polymeric hybrid NPs	Docetaxel + siRNA targeting Sonic Hedgehog gene	Metastatic prostate cancer	<i>In vitro/in vivo</i> (preclinical)	181
Gold nanorods inside silica nanoparticles (Au@MSNs) (photothermal)	Gold nanorod-silica NPs (photothermal)	Zoledronic acid	Metastatic breast cancer	<i>In vitro/in vivo</i> (preclinical)	182
Poly(lactone-co-β-amino ester)-AMID3100- <i>proMel</i>	Peptide-based NPs	AMID3100 + pro-Melittin	Metastatic breast cancer	<i>In vitro/in vivo</i> (preclinical)	183
PDGFRβ-targeted polymeric nanoparticle (cyclic peptide-modified)	Surface-modified polymeric NPs	siRNA (CAF-targeting)	Hepatocellular carcinoma	<i>In vitro/in vivo</i> (preclinical)	184
Cyanine dye-loaded nanoparticle-based PTT	Micelles (photothermal)	Pt(IV) prodrug + cyanine dye (Cypate)	Lung cancer	<i>In vitro/in vivo</i> (preclinical)	191
RGD-functionalized CuS NPs	Copper sulfide NPs	None (PTT agent)	Metastatic breast cancer	<i>In vitro/in vivo</i> (preclinical)	194
FIC-PDT	Photosensitizer NPs (PDT therapy)	Rho-kinase (ROCK) inhibitor + anti-PD-L1 antibody	Uveal melanoma	<i>In vitro/in vivo</i> (preclinical)	196
HA-modified MSN	HA-modified mesoporous silica NPs (MSN-HAS)	siRNA against TWIST1	Ovarian cancer	<i>In vitro/in vivo</i> (preclinical)	198
Carbon-based quantum dots (CQDs)/Cu ₂ O	Carbon quantum dot/Cu ₂ O hybrid NPs	None	Ovarian cancer	<i>In vitro</i> (preclinical)	203
(Tan-ZnO) quantum dots	Tangeretin-zinc oxide quantum dots	Tangeretin	Metastatic lung cancer	<i>In vitro/in vivo</i> (preclinical)	201



Table 2 Clinically approved nanomedicines for metastatic cancer treatment

Drug name	Drug	Nanocarrier	Metastatic cancer type	Regulatory status	Ref.
Doxil/Caelyx	Doxorubicin	PEGylated liposomes	Metastatic breast cancer	FDA-(Doxil) and EMA-(Caelyx) approved	204–206
Myocet	Doxorubicin	Liposomes	Metastatic breast cancer	EMA-approved	207 and 208
Lipusu	Paclitaxel	Liposomes	Advanced gastric cancer ^a	NMPA-approved	209
Genexol PM	Paclitaxel	Polymeric micelles	Metastatic breast cancer	Approved in South Korea	210 and 211
Onivyde	Irinotecan	PEGylated liposomes	Metastatic pancreatic adenocarcinoma	FDA- and EMA-approved	212 and 213
Pazenir	Paclitaxel	Polymeric micelles	Metastatic breast cancer, metastatic pancreatic cancer	Approved in South Korea	214 and 215
Abraxane	Paclitaxel	Albumin	Metastatic adenocarcinoma of the pancreas, metastatic breast cancer	FDA- and EMA-approved	216
DaunoXome	Daunorubicin	Liposomes	Advanced Kaposi's sarcoma ^a	FDA-approved	217
DepoCyt	Cytarabine	Liposomes	Lymphomatous meningitis ^b	FDA-approved, EMA-approved (withdrawn)	218

^a Metastatic disease not explicitly specified in regulatory approval. ^b Lymphomatous meningitis represents CNS dissemination of hematological malignancies and is functionally analogous to leptomeningeal metastases in solid tumors; therefore, it is included as a clinically relevant model of disseminated (metastasis-like) disease.

occurs *via* clathrin-, and caveolae-dependent endocytosis, or *via* micropinocytosis. However, following internalization, the NPs often became trapped in the endosomal-lysosomal stage, in which the acidic environment often degrades or destabilizes NPs. Endocytic uptake also often leads to perivascular accumulation, very limited tissue penetration, and, in general, heterogeneous intratumoral distribution of the drug. Efficient cargo release from endosomes into the cytosol remains the main limiting step in intracellular drug delivery and a critical barrier blocking the NPs treatment efficiency. Transcytosis can overcome the limits of endocytic uptake by actively transporting nanoparticles across endothelial cells, preventing perivascular trapping and enabling deeper, more uniform penetration into tumor tissue. By transporting particles through cells rather than leaving them stuck near vessels, transcytosis overcomes poor vascular permeability and dense stroma that limit endocytic delivery. Transcytosis is an ATP-dependent, active form of transport that is independent of vascular leakiness. It starts with endocytosis *via* adsorptive-mediated transcytosis (AMT) proposed by electrostatic interaction between anionic plasma membrane and cationic molecules, or receptor-mediated transcytosis (RMT) initiated by binding of the ligands to target receptors (Fig. 3). One of the next-generation strategies is the induction of transcytosis in cancer cells or epithelial cells to enable active extravasation and infiltration into the tumor, resulting in increased anticancer efficacy.²¹⁹ Several studies have demonstrated the potential of transcytosis-based nanoparticle strategies for deep tumor penetration. Cationized polymer–drug NPs were shown to possess strong active transcytosis ability, leading to effective suppression of established tumors reaching approximately 500 mm³ in volume.²²⁰ Similarly, another study demonstrated that positively charged poly(L-lysine)-functionalized upconversion NPs could be transported into deep tissues through transcytosis.²²¹ Despite these advances, strategies that actively promote transcytosis within solid tumors remain largely unexplored.

Zwitterionic polymer–drug conjugate PBEAGA-CPT exhibits an overall neutral surface charge and therefore circulates through blood without significant clearance. Upon contact with

endothelial/cancer cells, the overexpressed γ -glutamyl transpeptidase (GGT) removes the γ -glutamyls from PPBEAGA, generating primary amines and therefore resulting in a positive surface charge. This cationization allows membrane adsorption, rapid caveolae-mediated endocytosis, and subsequent transcytosis, enabling transendothelial extravasation and intra-tumoral infiltration. Consequently, enhanced antitumor activity and eradication of approximately 100 mm³ tumors as well as larger, exponentially growing tumors was observed.²²² In another study, enamine N-oxide-modified nanoparticles were developed, encapsulating a stearic acid-modified gemcitabine prodrug (GemC18) and a pSMAD2/3 inhibitor, galunisertib, for the treatment of pancreatic ductal adenocarcinoma. Upon reaching the tumor site, particles respond to the acidic TME and hypoxia, resulting in surface charge conversion to the positive state, promoting tumor penetration and nanoparticle disintegration, releasing the cargo.²²³ In a recent study, the RMT strategy of active transcytosis was demonstrated by metallic iron-oxide NPs aiming at overexpressed glucose transporters, mainly GLUT1, on glioblastoma endothelial cells. These glucuronic acid-functionalized NPs enhance GLUT-mediated transcytosis, which enables highly precise targeting of glioblastoma and overcoming the blood–brain barrier.²²⁴

Another approach involves cell-mediated nanoparticle delivery systems, which utilize living cells, such as immune cells, stem cells, or engineered cell carriers, to transport therapeutic nanoparticles directly to tumors. Because these cells naturally home to sites of inflammation, hypoxia, and cancer, they can overcome biological barriers that synthetic nanoparticles often fail to penetrate. This approach enhances targeted delivery, improves penetration into deep tumor regions, and reduces off-target toxicity. By combining the programmable behavior of cells with the tunable properties of nanoparticles, cell-mediated delivery represents a highly adaptable platform for precision nanomedicine, enabling more selective, efficient, and personalized cancer therapy.²²⁵

The physicochemical properties of NPs, mainly the shape, size, and stiffness, influence the specific uptake pathway involved in internalization. While clathrin- and caveolae-



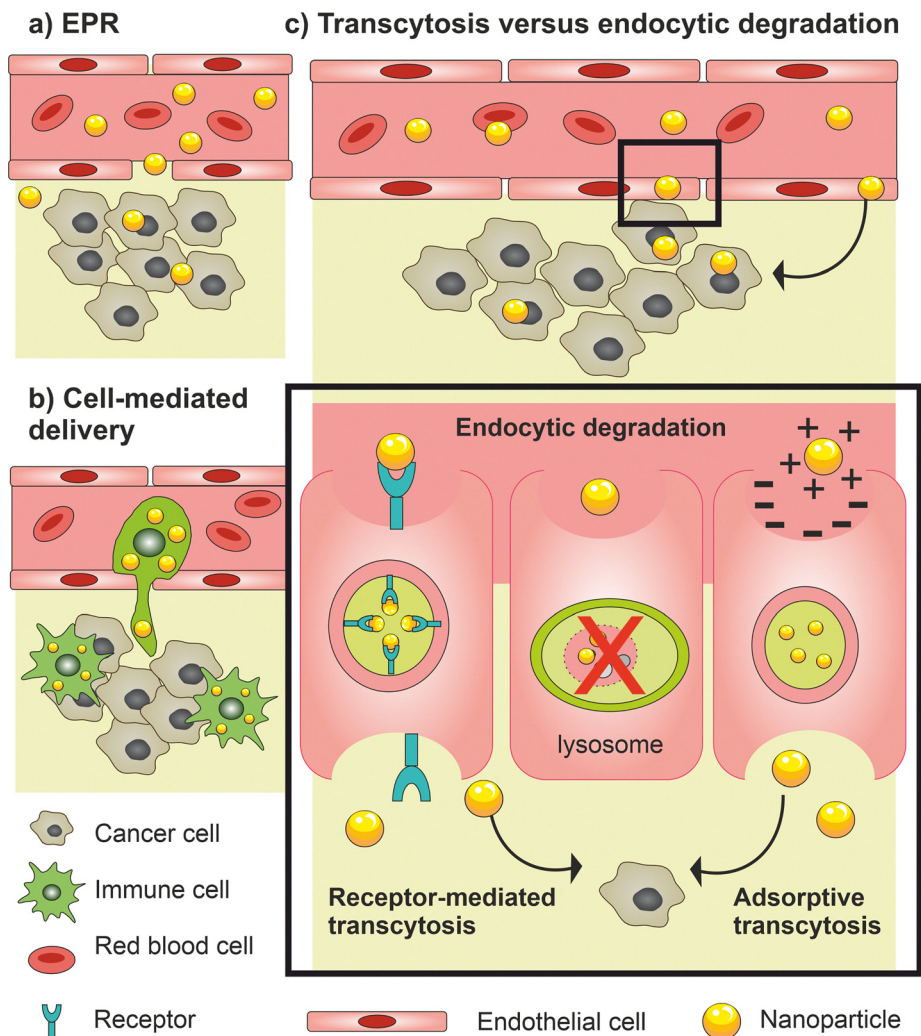


Fig. 3 Mechanisms for *in vivo* nanoparticle delivery. (a) Passive nanoparticle accumulation via the enhanced permeability and retention (EPR) effect, driven by leaky or fenestrated tumor vasculature and impaired lymphatic drainage; (b) cell-mediated delivery, whereby engineered immune cells internalize nanoparticles and actively migrate into tumors; (c) active transcytosis, in which nanoparticles are internalized by endothelial cells, trafficked across the cytoplasm, and released on the abluminal side, enabling transport across the vascular barrier while avoiding lysosomal endocytic degradation and perivascular retention. Adsorptive-mediated transcytosis (AMT) is driven by nonspecific electrostatic interactions between cationic nanoparticles and the anionic endothelial plasma membrane, resulting in relatively high uptake but limited selectivity and potential off-target transport. In contrast, receptor-mediated transcytosis (RMT) is initiated by specific ligand–receptor interactions, enabling controlled, cell-type-selective transport across the endothelium with improved targeting efficiency and reduced nonspecific uptake. Adapted from Pandit *et al.*²¹⁹

mediated endocytosis occur at comparable levels in cancer and healthy cells, macropinocytosis is markedly upregulated in cancer. This increase is driven by oncogenic alterations, most notably KRAS mutations, and is particularly prominent in highly aggressive malignancies such as breast, lung, and pancreatic cancers, where it supports enhanced nutrient uptake. This suggests that more malignant types of cancer exhibit elevated activity in macropinocytosis to sustain their biosynthetic and energetic needs of rapid proliferation and invasion. This uptake preference can be exploited in the design of nanotherapeutics that selectively target macropinocytosis. Moreover, given that cancer cells exhibit an overall negatively charged plasma membrane and are softer, charge-switching NPs have been developed to target the negatively charged

membrane by switching from negative charge to positive to specifically target cancer cells and increase intracellular internalization.²²⁶

The physicochemical properties favoring macropinocytosis include NP sizes typically larger than 200 nm, since these are too big to be internalized by the clathrin- or caveolae-dependent pathway, rod-shaped like NPs, due to an increase in membrane contact area and induction of asymmetric membrane wrapping, and lipoprotein-coated NPs that mimic nutrient-like cargo. These findings suggest that tuning NPs for intracellular uptake *via* transcytosis or macropinocytotic endocytosis may present a promising strategy to enhance transport across biological barriers and improve selective targeting of tumor cells.^{221,227}



6. Cargo-free nanotherapeutics

In nanomedicine, nanotherapeutics can be either formulated as a nanocarrier containing a conventional or novel drug as cargo to transport the treatment directly to the tumor site, or the nanoformulation itself can be an anticancer therapeutic. Drug-free unmodified gold nanoparticles (AuNPs) were found to suppress tumor cell proliferation through inhibition of MAPK signaling while also reversing EMT by disrupting the function of heparin-binding growth factors (HB-GFs), which are essential for angiogenesis and EMT progression. The underlying mechanisms included upregulation of *E*-cadherin expression and reduced expression of EMT-associated proteins such as Snail, *N*-cadherin, and vimentin. These observations were validated in two distinct orthotopic mouse models of ovarian cancer.²²⁸ Other types of metal NPs are non-persistent gold/copper ultrasmall-in-nano architectures (NAs). Evaluation of NAs in the chorioallantoic membrane (CAM) model of pancreatic ductal adenocarcinoma revealed their ability to modulate antimetastatic behavior, mainly through alterations in EMT-related protein and gene expression.²²⁹ Poly(ethyleneglycol)-functionalized dendrimers have shown to deplete bioavailable copper and therefore inhibit angiogenesis *in vivo*. In comparison to conventional chemotherapeutics, the dendrimer exhibited antitumor activity in a non-cytotoxic way, yet resulting in suppressed tumor proliferation and metastasis.²³⁰ In another study, mesoporous silica NPs (MSN-PEG/TA 25) have shown antimetastatic activity in breast cancer mouse models. Combined treatment with MSN-PEG/TA 25 and liposomal doxorubicin significantly improved survival in mice compared with treatment using liposomal doxorubicin alone. This approach may represent a more effective strategy to inhibit metastasis by targeting focal adhesion kinase (FAK) rather than EMT directly, as it suppresses cancer cell migration and interferes with several stages of the metastatic cascade, including invasion and colonization.²³¹ Other cargo-free nanotherapeutics with antimetastatic activity and potent synergic effect with conventional drugs are carbon nanotubes mimicking cytoskeletal filaments. They showed intrinsic anticancer activity and inhibition of metastasis *via* VEGF receptor targeting. In a murine metastatic melanoma model, treatment with these nanotubes reduced pulmonary metastases by more than 80%. When combined with paclitaxel, this antimetastatic effect increased to approximately 90%.²³² In immunocompetent 4T1 mice, intragastric administration of citrate- and PEG-coated silver nanoparticles significantly reduced lung metastases without affecting primary tumor growth or EMT-related gene expression. The observed effects instead correlated with modulation of inflammation-related pathways.²³³ A very recent study demonstrated that boron-incorporated alginate carbon nanogels significantly reduced lung metastases by over 85%. These particles selectively target TNBC cells by interaction with sialic acid on the cancer cells surface and result in disruption of F-actin and ROS-mediated cell cycle arrest.²³⁴ CXCR4-targeted metallo-nanodrugs were designed to treat breast cancer metastasis by inducing *in situ* oxidative stress enhanced by hypothermia and

by blocking the CXCR4/CXCL12 signaling axis. This approach inhibited invasion and migration of 4T1 cancer cells. In a BALB/c mouse mammary tumor model, these nanostructures suppressed both primary tumor growth and lung metastases with minimal side effects, demonstrating strong potential for precision nanomedicine.²³⁵ Targeting cancer metastasis does not necessarily need to focus solely on cancer cells but can also involve modulation of the complex tumor microenvironment. For example, gold-core silver-shell nanoparticles (Au@Ag) have been reported to affect cancer-promoting activity of CAFs by altering their communication with tumor cells. Transcriptomic analysis revealed alterations in the secretory profiles of nanoparticle-exposed CAFs, especially downregulation of *Spp1* expression observed in the 4T1 *in vivo* model following Au@Ag treatment. Elevated expression of this protein has been associated to poor survival in breast cancer patients, suggesting that it may represent a promising target for future therapeutic strategies.²³¹ Overall, these findings demonstrate that rationally engineered nanoparticles can function as standalone antimetastatic agents or synergize with conventional drugs by modulating metastasis-related pathways or directly inhibiting tumor progression.

7. Challenges and future perspectives

Owing to their structural complexity and the need for multi-functional design, including optimized drug encapsulation, controlled release, tissue penetration, biodistribution, metabolism, and clearance, the development and comprehensive evaluation of nanotherapeutics remain highly challenging. Many factors complicate formulation design, pharmacokinetic, as well as pharmacodynamic characterization, and hinder the accurate assessment of therapeutic efficacy and safety. In addition, nanomedicine delivery is impeded by barriers within the tumor microenvironment, which constitutes a highly complex, heterogeneous, and patient-specific network of interactions. At the same time, nanotherapeutics themselves may elicit toxicity or unintended immune responses within the local tissue microenvironment. Taken together, these challenges underscore the need for more rational nanotherapeutic design, improved preclinical models, and integrative evaluation strategies. A major factor contributing to impaired nanomedicine delivery is the compromised EPR effect within the tumor microenvironment. The EPR effect varies substantially depending on the xenograft mouse model used and shows pronounced heterogeneity across patients, limiting its reliability for effective nanoparticle accumulation.^{236,237} The EPR effect also exhibits substantial variability between xenograft mouse models and patients. Human tissues display markedly higher blood flow rates than rodent tissues, resulting in greater shear forces and faster vascular washout. This may partially explain the superior accumulation of nanomedicines in rodent tumors and pose a significant challenge to the clinical translatability of preclinical nanomedicine findings.¹⁴⁷ Furthermore, differing tumor vasculature and desmoplasia restrain the proper delivery of



Table 3 Nanoparticle-based therapies undergoing clinical evaluation for metastatic indications

Drug/system name	Cargo	Nanocarrier	Metastatic cancer type	Phase	NCT identifier
AGuIX (NH TherAguix)	NA	Gadolinium-chelated polysiloxane based NPs	Advanced/unresectable pancreatic cancer ^a	Phase I/II	NCT04789486
AGuIX (NH TherAguix)	NA	Gadolinium-chelated polysiloxane based NPs	Multiple brain metastases	Phase II	NCT03818386
BIND-014	Docetaxel	PEG-PLGA NPs	Metastatic castration-resistant prostate cancer	Phase II	NCT01812746
NanoTherm	None	Aminosilane-coated iron oxide NPs	Glioblastoma (recurrent) ^a	NA	NCT06271421
Lipo-MERIT vaccine	Tumor-associated antigen mRNA (immunotherapy)	Cationic lipoplex (DOTMA/DOPE)	Advanced melanoma ^a	Phase I	NCT02410733
CriPec [®] , CPC634	Docetaxel	PEG-polymeric NPs	Advanced solid malignancies ^a	Phase I	NCT02442531
64Cu-MM-302	Doxorubicin	PEGylated liposomes	HER2 positive metastatic breast cancer	Phase I	NCT01304797
PRECIOUS-01	Threitolceramide-6 + NY-ESO-1 cancer-testis antigen peptides	Poly(lactic-co-glycolic acid) (PLGA) NPs	Advanced NY-ESO-1-positive cancers ^a	Phase I	NCT04751786
ThermoDOX [®]	Doxorubicin	Lyso-thermosensitive liposomes	Liver metastases	Phase I	NCT02181075
Docetaxel-PM	Docetaxel	Polymeric micelles	Metastatic esophageal squamous cell carcinoma	Phase II	NCT03585673
Docetaxel-PNP	Docetaxel	Liposomes	Advanced solid malignancies ^a	Phase I	NCT01103791
LipoVNB	Vinorelbine tartrate	Liposomes	Advanced malignancy ^a	Phase I, II	NCT02925000
iExosomes	KRAS G12D siRNA	Exosomes	Metastatic pancreas cancer with KrasG12D mutation	Phase I, II	NCT03608631
Sarah Nanotechnology System	NA	Iron oxide nanoparticles	Advanced metastatic solid tumors (stage 4)	NA	NCT07224464
NBTXR3 (Hensify)	Anti-PD-1	Hafnium oxide nanoparticles	Metastatic non-small cell lung cancer, liver metastases	Phase I	NCT03589339
NBTXR3 (Hensify)	Anti-PD-1	Hafnium oxide nanoparticles	Advanced solid malignancies ^a	Phase I, II	NCT05039632
NBTXR3 (Hensify)	Anti-PD-1	Hafnium oxide nanoparticles	Advanced or borderline-resectable pancreatic cancer ^a	Phase I	NCT04484909
E-EDV-D682	PNU-159682	EnGeneIC Dream Vector (EDV) nanocells	Metastatic pancreatic cancer	Phase I, II	NCT07049055
Carbon nanoparticle-loaded iron [CNSI-Fe(II)]	Ferrous iron (Fe ²⁺)	Carbon-based nanoparticles	Advanced solid tumors ^a	Phase I	NCT06048367
CPX-1	Irinotecan floxuridine	Liposomes	Advanced/metastatic colorectal cancer	Phase II	NCT00361842
Liposomal irinotecan	Irinotecan	Liposomes	Metastatic pancreatic, colorectal, gastroesophageal, or biliary cancer	Phase I, II	NCT03337087
FF-10850	Topotecan	Liposomes	Advanced solid cancers ^a	Phase I	NCT04047251

^a Metastatic disease not explicitly specified in regulatory approval.

nanotherapeutics to tumor sites. On top of this, in clinical practice, nanotherapeutics are frequently administered in later treatment lines or in combination with other anticancer agents, and prior or concomitant therapies may significantly remodel tumor vasculature and stromal architecture, thereby further modulating the EPR effect. In metastasis, the EPR effect differs even more.²³⁸ In macrometastasis, which are highly evolved and angiogenic, the EPR effect may be present; however, micrometastasis may lack any vasculature. Depending on the secondary site, each tissue where metastasis is present constitutes another barrier for nanotherapeutics, often different from the one present in the primary tumor. In brain metastases, the blood-brain barrier is highly restrictive and has very limited permeability. In bone metastasis, the complex dense matrix limits the diffusion of NPs into lesions, and hydroxyapatite can bind charged NPs non-specifically. In the liver, NPs must sustain clearance by Kupfer cells and avoid accumulation in sinusoidal spaces. One of the solutions may be the

stratification of patients before the nanotherapeutic delivery by EPR-predictive biomarkers or by imaging agents.²³⁹ Patients with dense, non-leaky tumor vasculature may benefit from active or bypass delivery strategies, including targeting of the tumor vasculature, locoregional administration, integrin-RGD-mediated targeting, or cell-mediated delivery approaches. However, each of these potential therapies comes with many other obstacles as well. The phenomenon known as “biomolecular corona” is a significant obstacle occurring when nanoparticles meet biological fluids. During this interaction, biomolecules adsorb onto the nanoparticle surface, forming a corona that can mask targeting ligands and alter the physicochemical properties of the nanocarrier. In addition, the biomolecular corona may trigger immune recognition, leading to rapid clearance by the mononuclear phagocyte system before the nanoparticles reach their intended target.^{240,241} When the NPs successfully reach near the tumor site, another physiological barrier is present called interstitial fluid pressure (IFP).



Whereas in normal tissues the IFP is low, in solid primary tumors and macrometastases, there is a significant elevation in IFP due to leaky vessels and dense matrix.²⁴² This IFP elevation causes limited extravasation of NPs and their accumulation on the tumor site periphery. So even if the EPR effect favors the successful transport of NPs into the tumor site, the high IFP reduces it dramatically. As well as EPR, the IFP varies through different stages of metastatic formation. While micrometastases show almost no IFP, the fully developed macrometastases mimic the solid primary tumor IFP. Some nanodrugs were also found to be accumulating in the body, mainly in the liver and spleen, for a long time.^{243,244} Their improper elimination then results in physical damage of the affected tissue, resulting in inflammation and toxic reactions, including the production of ROS and elevation in pro-inflammatory cytokines.^{245,246} Therefore, there is a strong interest in the development of biodegradable or biocompatible nanocarriers. RGD-, CD44-, and transferrin-targeted nanotherapies, as mentioned above, represent promising treatment strategies for targeting cancer cells; however, their efficacy in a metastasis setting is heavily dependent on intra- and inter-metastatic heterogeneity.²⁴⁷⁻²⁴⁹ The expression of these target molecules is highly variable across different stages of the disease and metastatic organ sites and also within the same lesion itself. Expression of RGD-binding integrins is often reduced in dormant and poorly angiogenic micrometastases. CD44 expression exhibits temporal and spatial heterogeneity across metastatic lesions and undergoes switching of isoforms (CD44v ↔ CD44s) during EMT.²⁵⁰ Transferrin receptor expression is generally higher in rapidly growing tumor sites due to increased iron demand; however in slow-growing and dormant lesions the expression is lower, contributing to the overall heterogeneity between primary and metastatic disease. The translational phase of nanomedicine also presents additional challenges due to a lack of validated analytical methods for biodistribution and pharmacokinetics, and a lack of predictive biological models. Because of these translational challenges, the current approval rate of nanotherapeutics in oncology is estimated to be below 10%, and in the Phase III trials, the failure rate is up to 86%.²⁵¹ However, these percentages are broadly comparable to those of conventional cancer therapies, reflecting the overall complexity of cancer drug development. In recent years, the rapid advancement of artificial intelligence (AI) has introduced powerful tools for nanomedicine development, ranging from the rational design of nanoparticles with improved delivery efficiency to decision-support systems that help identify which nanotherapeutics are most likely to succeed in clinical trials. The gap between early-stage nanoparticle development and large-scale translation processes could be bridged by these novel tools, facilitating the successful clinical translation of nanoparticle therapeutics²⁵² (Table 3).

8. Conclusions

While primary tumors can often be removed surgically, metastatic disease presents a far greater challenge. Once cancer has

spread to distant organs, it can become incurable, significantly reducing patient survival rates. Recent advances in antimetastatic approaches, such as migrastatic therapies, nanomedicine, and targeted drug delivery, offer promising opportunities to improve patient outcomes. These therapies focus on disrupting the key mechanisms that drive metastasis, such as neovascularisation, cancer cell motility, metabolic and phenotypic plasticity, and immune evasion, providing a more precise and effective approach to cancer treatment. By targeting the critical steps of the metastatic cascade, modifying the tumor microenvironment, and integrating them into multimodal treatment strategies, antimetastatic therapies may improve patient survival, quality of life, and overall cancer care for patients worldwide. NPs have emerged as a powerful tool in cancer therapy, offering unique physicochemical properties and versatile functionalities. NPs are particularly promising due to their ability to target metastatic processes at the cellular and molecular levels. The versatility of NPs enables precise delivery of therapeutic agents, reduction of systemic toxicity, and modulation of the tumor microenvironment. Additionally, NPs can also be integrated into photodynamic therapy (PDT) and photothermal therapy (PTT) strategies. While preclinical studies demonstrate the potential of NPs in antimetastatic therapy, challenges remain in translating these findings into clinical practice. Key barriers include large-scale production, long-term biocompatibility, and precise targeting. Ongoing research aims to address these limitations, with advances in NP design, surface functionalization, and safety profiles bringing nanoparticle-based antimetastatic treatments closer to clinical reality.

Author contributions

TM wrote the article, MF and MR conceived the structure and created images, and MF, MM, and MR revised the article.

Conflicts of interest

The authors declare no conflicts of interest.

Data availability

No new data were generated or analyzed in this study.

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References

- H. Dillekås, M. S. Rogers and O. Straume, Are 90% of deaths from cancer caused by metastases?, *Cancer Med.*, 2019, **8**(12), 5574–5576, DOI: [10.1002/cam4.2474](https://doi.org/10.1002/cam4.2474).
- D. Hanahan and R. A. Weinberg, Hallmarks of Cancer: The Next Generation, *Cell*, 2011, **144**(5), 646–674, DOI: [10.1016/j.cell.2011.02.013](https://doi.org/10.1016/j.cell.2011.02.013).
- M. L. Costa, I. de Andrade Rosa, L. Andrade, C. Mermelstein and C. C. Coutinho, Distinct interactions between epithelial and mesenchymal cells control cell morphology and collective migration during sponge epithelial to mesenchymal transition, *J. Morphol.*, 2020, **281**(2), 183–195, DOI: [10.1002/jmor.21090](https://doi.org/10.1002/jmor.21090).
- K. K. Youssef and M. A. Nieto, Epithelial–mesenchymal transition in tissue repair and degeneration, *Nat. Rev. Mol. Cell Biol.*, 2024, 1–20, DOI: [10.1038/s41580-024-00733-z](https://doi.org/10.1038/s41580-024-00733-z).
- M. A. Nieto, R. Y.-J. Huang, R. A. Jackson and J. P. Thiery, EMT: 2016, *Cell*, 2016, **166**(1), 21–45, DOI: [10.1016/j.cell.2016.06.028](https://doi.org/10.1016/j.cell.2016.06.028).
- E. D'Angelo, *et al.*, Intrinsic and Extrinsic Modulators of the Epithelial to Mesenchymal Transition: Driving the Fate of Tumor Microenvironment, *Front. Oncol.*, 2020, **10**, 1122, DOI: [10.3389/fonc.2020.01122](https://doi.org/10.3389/fonc.2020.01122).
- H. Goetz, J. R. Melendez-Alvarez, L. Chen and X.-J. Tian, A plausible accelerating function of intermediate states in cancer metastasis, *PLoS Comput. Biol.*, 2020, **16**(3), e1007682, DOI: [10.1371/journal.pcbi.1007682](https://doi.org/10.1371/journal.pcbi.1007682).
- F. Junaid, G. Tomic, R. Kemp and D. J. Winton, Single-copy Snail upregulation causes partial epithelial–mesenchymal transition in colon cancer cells, *BMC Cancer*, 2023, **23**(1), 153, DOI: [10.1186/s12885-023-10581-3](https://doi.org/10.1186/s12885-023-10581-3).
- A. E. de Groot, S. Roy, J. S. Brown, K. J. Pienta and S. R. Amend, Revisiting Seed and Soil: Examining the Primary Tumor and Cancer Cell Foraging in Metastasis, *Mol. Cancer Res.*, 2017, **15**(4), 361–370, DOI: [10.1158/1541-7786.MCR-16-0436](https://doi.org/10.1158/1541-7786.MCR-16-0436).
- Y. Zhu, M. Luo, M. Brooks, S. G. Clouthier and M. S. Wicha, Biological and clinical significance of cancer stem cell plasticity, *Clin. Transl. Med.*, 2014, **3**(1), 32, DOI: [10.1186/s40169-014-0032-3](https://doi.org/10.1186/s40169-014-0032-3).
- O. N. Gofrit, *et al.*, Patterns of metastases progression – The linear parallel ratio, *PLoS One*, 2022, **17**(9), e0274942, DOI: [10.1371/journal.pone.0274942](https://doi.org/10.1371/journal.pone.0274942).
- E. Bugyik, *et al.*, Mechanisms of vascularization in murine models of primary and metastatic tumor growth, *Chin. J. Cancer*, 2016, **35**(1), 19, DOI: [10.1186/s40880-016-0083-5](https://doi.org/10.1186/s40880-016-0083-5).
- Y. Zhao, *et al.*, ROS impairs tumor vasculature normalization through an endocytosis effect of caveolae on extracellular SPARC, *Cancer Cell Int.*, 2023, **23**(1), 152, DOI: [10.1186/s12935-023-03003-8](https://doi.org/10.1186/s12935-023-03003-8).
- G. Kaur and B. Roy, Oding Tumor Angiogenesis for Therapeutic Advancements: Mechanistic Insights, *Biomedicines*, 2024, **12**(4), 827, DOI: [10.3390/biomedicines12040827](https://doi.org/10.3390/biomedicines12040827).
- V. M. Asprițoiu, I. Stoica, C. Bleotu and C. C. Diaconu, Epigenetic Regulation of Angiogenesis in Development and Tumors Progression: Potential Implications for Cancer Treatment, *Front. Cell Dev. Biol.*, 2021, **9**, 689962, DOI: [10.3389/fcell.2021.689962](https://doi.org/10.3389/fcell.2021.689962).
- F. Lopes-Coelho, F. Martins, S. A. Pereira and J. Serpa, Anti-Angiogenic Therapy: Current Challenges and Future Perspectives, *Int. J. Mol. Sci.*, 2021, **22**(7), 3765, DOI: [10.3390/ijms22073765](https://doi.org/10.3390/ijms22073765).
- L. Wang, W.-Q. Liu, S. Broussy, B. Han and H. Fang, Recent advances of anti-angiogenic inhibitors targeting VEGF/VEGFR axis, *Front. Pharmacol.*, 2024, **14**, 1307860, DOI: [10.3389/fphar.2023.1307860](https://doi.org/10.3389/fphar.2023.1307860).
- M. S. Pepper, Lymphangiogenesis and Tumor Metastasis: Myth or Reality?, *Clin. Cancer Res. Off. J. Am. Assoc. Cancer Res.*, 2001, **7**, 462–468.
- D. Jones, E. R. Pereira and T. P. Padera, Growth and Immune Evasion of Lymph Node Metastasis, *Front. Oncol.*, 2018, **8**, 36, DOI: [10.3389/fonc.2018.00036](https://doi.org/10.3389/fonc.2018.00036).
- B. Garmy-Susini, *et al.*, PI3K α activates integrin $\alpha 4\beta 1$ to establish a metastatic niche in lymph nodes, *Proc. Natl. Acad. Sci. U. S. A.*, 2013, **110**(22), 9042–9047, DOI: [10.1073/pnas.1219603110](https://doi.org/10.1073/pnas.1219603110).
- W.-F. Wei, *et al.*, Periostin + cancer-associated fibroblasts promote lymph node metastasis by impairing the lymphatic endothelial barriers in cervical squamous cell carcinoma, *Mol. Oncol.*, 2021, **15**(1), 210–227, DOI: [10.1002/1878-0261.12837](https://doi.org/10.1002/1878-0261.12837).
- C. D. Commerford, *et al.*, Mechanisms of Tumor-Induced Lymphovascular Niche Formation in Draining Lymph Nodes, *Cell Rep.*, 2018, **25**(13), 3554–3563.e4, DOI: [10.1016/j.celrep.2018.12.002](https://doi.org/10.1016/j.celrep.2018.12.002).
- X. Wang, *et al.*, Detection of circulating tumor cells in human peripheral blood using surface-enhanced Raman scattering nanoparticles, *Cancer Res.*, 2011, **71**(5), 1526–1532, DOI: [10.1158/0008-5472.CAN-10-3069](https://doi.org/10.1158/0008-5472.CAN-10-3069).
- M. Giuliano, *et al.*, Perspective on Circulating Tumor Cell Clusters: Why It Takes a Village to Metastasize, *Cancer Res.*, 2018, **78**(4), 845–852, DOI: [10.1158/0008-5472.CAN-17-2748](https://doi.org/10.1158/0008-5472.CAN-17-2748).
- H. Zhang, *et al.*, Detection Methods and Clinical Applications of Circulating Tumor Cells in Breast Cancer, *Front. Oncol.*, 2021, **11**, 652253, DOI: [10.3389/fonc.2021.652253](https://doi.org/10.3389/fonc.2021.652253).
- J. Park, *et al.*, c-MET-positive circulating tumor cells and cell-free DNA as independent prognostic factors in hormone receptor-positive/HER2-negative metastatic breast cancer, *Breast Cancer Res.*, 2024, **26**(1), 13, DOI: [10.1186/s13058-024-01768-y](https://doi.org/10.1186/s13058-024-01768-y).
- P. Salu and K. M. Reindl, Advancements in Circulating Tumor Cell Research: Bridging Biology and Clinical Applications, *Cancers*, 2024, **16**(6), 1213, DOI: [10.3390/cancers16061213](https://doi.org/10.3390/cancers16061213).
- K. Pantel and C. Alix-Panabières, Crucial roles of circulating tumor cells in the metastatic cascade and tumor immune escape: biology and clinical translation, *J. Immunother. Cancer*, 2022, **10**(12), e005615, DOI: [10.1136/jitc-2022-005615](https://doi.org/10.1136/jitc-2022-005615).
- K. Kurma and C. Alix-Panabières, Mechanobiology and survival strategies of circulating tumor cells: a process



- towards the invasive and metastatic phenotype, *Front. Cell Dev. Biol.*, 2023, **11**, 1188499, DOI: [10.3389/fcell.2023.1188499](https://doi.org/10.3389/fcell.2023.1188499).
- 30 L. Wang, *et al.*, Circulating galectin-3 promotes tumor-endothelium-adhesion by upregulating ICAM-1 in endothelium-derived extracellular vesicles, *Front. Pharmacol.*, 2022, **13**, 979474, DOI: [10.3389/fphar.2022.979474](https://doi.org/10.3389/fphar.2022.979474).
- 31 J. A. Espina, M. H. Cordeiro, M. Milivojevic, I. Pajić-Lijaković and E. H. Barriga, Response of cells and tissues to shear stress, *J. Cell Sci.*, 2023, **136**(18), jcs260985, DOI: [10.1242/jcs.260985](https://doi.org/10.1242/jcs.260985).
- 32 C. Garrido, *et al.*, MHC class I molecules act as tumor suppressor genes regulating the cell cycle gene expression, invasion and intrinsic tumorigenicity of melanoma cells, *Carcinogenesis*, 2012, **33**(3), 687–693, DOI: [10.1093/carcin/bgr318](https://doi.org/10.1093/carcin/bgr318).
- 33 Q. Zhan, *et al.*, New insights into the correlations between circulating tumor cells and target organ metastasis, *Signal. Transduction Targeted Ther.*, 2023, **8**(1), 1–23, DOI: [10.1038/s41392-023-01725-9](https://doi.org/10.1038/s41392-023-01725-9).
- 34 C. Qi, *et al.*, P-Selectin-Mediated Adhesion between Platelets and Tumor Cells Promotes Intestinal Tumorigenesis in ApcMin/+Mice, *Int. J. Biol. Sci.*, 2015, **11**(6), 679–687, DOI: [10.7150/ijbs.11589](https://doi.org/10.7150/ijbs.11589).
- 35 Y. Sun, *et al.*, Platelet-mediated circulating tumor cell evasion from natural killer cell killing through immune checkpoint CD155-TIGIT, *Hepatology*, 2025, **81**, 791–807, DOI: [10.1097/HEP.0000000000000934](https://doi.org/10.1097/HEP.0000000000000934).
- 36 S. Anvari, E. Osei and N. Maftoon, Interactions of platelets with circulating tumor cells contribute to cancer metastasis, *Sci. Rep.*, 2021, **11**(1), 15477, DOI: [10.1038/s41598-021-94735-y](https://doi.org/10.1038/s41598-021-94735-y).
- 37 M. Labelle, S. Begum and R. O. Hynes, Platelets guide the formation of early metastatic niches, *Proc. Natl. Acad. Sci. U. S. A.*, 2014, **111**(30), E3053–E3061, DOI: [10.1073/pnas.1411082111](https://doi.org/10.1073/pnas.1411082111).
- 38 S. Mao, *et al.*, Adhesion analysis of single circulating tumor cells on a base layer of endothelial cells using open microfluidics, *Chem. Sci.*, 2018, **9**(39), 7694–7699, DOI: [10.1039/C8SC03027H](https://doi.org/10.1039/C8SC03027H).
- 39 Y. Shamay, *et al.*, P-selectin is a nanotherapeutic delivery target in the tumor microenvironment, *Sci. Transl. Med.*, 2016, **8**(345), 345ra87, DOI: [10.1126/scitranslmed.aaf7374](https://doi.org/10.1126/scitranslmed.aaf7374).
- 40 J. Cools-Lartigue, *et al.*, Neutrophil extracellular traps sequester circulating tumor cells and promote metastasis, *J. Clin. Invest.*, 2013, **123**(8), 3446–3458, DOI: [10.1172/JCI67484](https://doi.org/10.1172/JCI67484).
- 41 J. Albregues, *et al.*, Neutrophil extracellular traps produced during inflammation awaken dormant cancer cells in mice, *Science*, 2018, **361**(6409), eaao4227, DOI: [10.1126/science.aao4227](https://doi.org/10.1126/science.aao4227).
- 42 Y. Dai, *et al.*, Anoikis resistance-protagonists of breast cancer cells survive and metastasize after ECM detachment, *Cell Commun. Signalling*, 2023, **21**(1), 190, DOI: [10.1186/s12964-023-01183-4](https://doi.org/10.1186/s12964-023-01183-4).
- 43 M. C. Guadamillas and A. Cerezo, and M. A. Del Pozo, Overcoming anoikis-pathways to anchorage-independent growth in cancer, *J. Cell Sci.*, 2011, **124**(Pt 19), 3189–3197, DOI: [10.1242/jcs.072165](https://doi.org/10.1242/jcs.072165).
- 44 X. Wen, *et al.*, Anoikis resistance regulates immune infiltration and drug sensitivity in clear-cell renal cell carcinoma: insights from multi omics, single cell analysis and in vitro experiment, *Front. Immunol.*, 2024, **15**, 1427475, DOI: [10.3389/fimmu.2024.1427475](https://doi.org/10.3389/fimmu.2024.1427475).
- 45 M. Monavarian, *et al.*, Development of adaptive anoikis resistance promotes metastasis that can be overcome by CDK8/19 Mediator kinase inhibition, *bioRxiv*, 2023, preprint, 2023.12.04.569970, DOI: [10.1101/2023.12.04.569970](https://doi.org/10.1101/2023.12.04.569970).
- 46 X. Cheng and K. Cheng, Visualizing cancer extravasation: from mechanistic studies to drug development, *Cancer Metastasis Rev.*, 2021, **40**(1), 71–88, DOI: [10.1007/s10555-020-09942-2](https://doi.org/10.1007/s10555-020-09942-2).
- 47 A. Dupas, J. G. Goetz and N. Osmani, Extravasation of immune and tumor cells from an endothelial perspective, *J. Cell Sci.*, 2024, **137**(21), jcs262066, DOI: [10.1242/jcs.262066](https://doi.org/10.1242/jcs.262066).
- 48 S. Di Russo, *et al.*, Beyond the barrier: the immune-inspired pathways of tumor extravasation, *Cell Commun. Signalling*, 2024, **22**(1), 104, DOI: [10.1186/s12964-023-01429-1](https://doi.org/10.1186/s12964-023-01429-1).
- 49 Y. Du, *et al.*, Metadherin regulates actin cytoskeletal remodeling and enhances human gastric cancer metastasis via epithelial-mesenchymal transition, *Int. J. Oncol.*, 2017, **51**(1), 63–74, DOI: [10.3892/ijo.2017.4002](https://doi.org/10.3892/ijo.2017.4002).
- 50 G. Hu, Y. Wei and Y. Kang, The multifaceted role of MTDH/AEG-1 in cancer progression, *Clin. Cancer Res.*, 2009, **15**(18), 5615–5620, DOI: [10.1158/1078-0432.CCR-09-0049](https://doi.org/10.1158/1078-0432.CCR-09-0049).
- 51 D. Ribatti and F. Pezzella, Vascular Co-Option and Other Alternative Modalities of Growth of Tumor Vasculature in Glioblastoma, *Front. Oncol.*, 2022, **12**, 874554, DOI: [10.3389/fonc.2022.874554](https://doi.org/10.3389/fonc.2022.874554).
- 52 N. Aceto, *et al.*, Circulating Tumor Cell Clusters are Oligoclonal Precursors of Breast Cancer Metastasis, *Cell*, 2014, **158**(5), 1110–1122, DOI: [10.1016/j.cell.2014.07.013](https://doi.org/10.1016/j.cell.2014.07.013).
- 53 P. Yang, Y. Hu and Q. Zhou, The CXCL12-CXCR4 Signaling Axis Plays a Key Role in Cancer Metastasis and is a Potential Target for Developing Novel Therapeutics against Metastatic Cancer, *Curr. Med. Chem.*, 2020, **27**(33), 5543–5561, DOI: [10.2174/09298673266661911131131110](https://doi.org/10.2174/09298673266661911131131110).
- 54 W. A. González-Arriagada, *et al.*, CCR5/CCL5 axis is linked to a poor outcome, and inhibition reduces metastasis in oral squamous cell carcinoma, *J. Cancer Res. Clin. Oncol.*, 2023, **149**(19), 17335–17346, DOI: [10.1007/s00432-023-05443-1](https://doi.org/10.1007/s00432-023-05443-1).
- 55 R. Huang and E. K. Rofstad, Integrins as therapeutic targets in the organ-specific metastasis of human malignant melanoma, *J. Exp. Clin. Cancer Res.*, 2018, **37**(1), 92, DOI: [10.1186/s13046-018-0763-x](https://doi.org/10.1186/s13046-018-0763-x).
- 56 S. Köhler, S. Ullrich, U. Richter and U. Schumacher, E-/P-selectins and colon carcinoma metastasis: first in vivo



- evidence for their crucial role in a clinically relevant model of spontaneous metastasis formation in the lung, *Br. J. Cancer*, 2010, **102**(3), 602–609, DOI: [10.1038/sj.bjc.6605492](https://doi.org/10.1038/sj.bjc.6605492).
- 57 B. Costa-Silva, *et al.*, Pancreatic cancer exosomes initiate pre-metastatic niche formation in the liver, *Nat. Cell Biol.*, 2015, **17**(6), 816–826, DOI: [10.1038/ncb3169](https://doi.org/10.1038/ncb3169).
- 58 Y. Wang, *et al.*, Pre-metastatic niche: formation, characteristics and therapeutic implication, *Signal. Transduction Targeted Ther.*, 2024, **9**(1), 236, DOI: [10.1038/s41392-024-01937-7](https://doi.org/10.1038/s41392-024-01937-7).
- 59 J. Jia, *et al.*, Neutrophils in the premetastatic niche: key functions and therapeutic directions, *Mol. Cancer*, 2024, **23**(1), 200, DOI: [10.1186/s12943-024-02107-7](https://doi.org/10.1186/s12943-024-02107-7).
- 60 V. Low, J. Blenis and A. P. Gomes, Targeting the premetastatic niche: epigenetic therapies in the spotlight, *Signal. Transduction Targeted Ther.*, 2020, **5**(1), 68, DOI: [10.1038/s41392-020-0165-3](https://doi.org/10.1038/s41392-020-0165-3).
- 61 R. R. Gomis and S. Gawrzak, Tumor cell dormancy, *Mol. Oncol.*, 2017, **11**(1), 62–78, DOI: [10.1016/j.molonc.2016.09.009](https://doi.org/10.1016/j.molonc.2016.09.009).
- 62 H. Endo and M. Inoue, Dormancy in cancer, *Cancer Sci.*, 2019, **110**(2), 474–480, DOI: [10.1111/cas.13917](https://doi.org/10.1111/cas.13917).
- 63 H. Wang, S. Wang, M. Huang, X. Liang, Y.-J. Tang and Y. Tang, Targeting Immune-Mediated Dormancy: A Promising Treatment of Cancer, *Front. Oncol.*, 2019, **9**, 498, DOI: [10.3389/fonc.2019.00498](https://doi.org/10.3389/fonc.2019.00498).
- 64 S. Bakhshandeh, C. Werner, P. Fratzl and A. Cipitria, Microenvironment-mediated cancer dormancy: insights from metastability theory, *Proc. Natl. Acad. Sci. U. S. A.*, 2022, **119**(1), e2111046118, DOI: [10.1073/pnas.2111046118](https://doi.org/10.1073/pnas.2111046118).
- 65 K. Santos-de-Frutos and N. Djouder, When dormancy fuels tumour relapse, *Commun. Biol.*, 2021, **4**(1), 747, DOI: [10.1038/s42003-021-02257-0](https://doi.org/10.1038/s42003-021-02257-0).
- 66 Z. R. Chalmers, *et al.*, Analysis of 100 000 human cancer genomes reveals the landscape of tumor mutational burden, *Genome Med.*, 2017, **9**(1), 34, DOI: [10.1186/s13073-017-0424-2](https://doi.org/10.1186/s13073-017-0424-2).
- 67 A. Ansari, S. K. Ray, M. Sharma, R. Rawal and P. Singh, Tumor Mutational Burden as a Biomarker of Immunotherapy Response: An Immunogram Approach in Oncoimmunology, *Curr. Mol. Med.*, 2024, **24**(12), 1461–1469, DOI: [10.2174/0115665240266906231024111920](https://doi.org/10.2174/0115665240266906231024111920).
- 68 N. A. Rizvi, *et al.*, Cancer immunology. Mutational landscape determines sensitivity to PD-1 blockade in non-small cell lung cancer, *Science*, 2015, **348**(6230), 124–128, DOI: [10.1126/science.aaa1348](https://doi.org/10.1126/science.aaa1348).
- 69 L. R. Yates, *et al.*, Subclonal diversification of primary breast cancer revealed by multiregion sequencing, *Nat. Med.*, 2015, **21**(7), 751–759, DOI: [10.1038/nm.3886](https://doi.org/10.1038/nm.3886).
- 70 B. Vogelstein, N. Papadopoulos, V. E. Velculescu, S. Zhou, L. A. Diaz and K. W. Kinzler, Cancer genome landscapes, *Science*, 2013, **339**(6127), 1546–1558, DOI: [10.1126/science.1235122](https://doi.org/10.1126/science.1235122).
- 71 G. Zhu, L. Pei, H. Xia, Q. Tang and F. Bi, Role of oncogenic KRAS in the prognosis, diagnosis and treatment of colorectal cancer, *Mol. Cancer*, 2021, **20**(1), 143, DOI: [10.1186/s12943-021-01441-4](https://doi.org/10.1186/s12943-021-01441-4).
- 72 M. P. Kim, *et al.*, Oncogenic KRAS Recruits an Expansive Transcriptional Network through Mutant p53 to Drive Pancreatic Cancer Metastasis, *Cancer Discovery*, 2021, **11**(8), 2094–2111, DOI: [10.1158/2159-8290.CD-20-1228](https://doi.org/10.1158/2159-8290.CD-20-1228).
- 73 T. J. Lynch, *et al.*, Activating mutations in the epidermal growth factor receptor underlying responsiveness of non-small-cell lung cancer to gefitinib, *N. Engl. J. Med.*, 2004, **350**(21), 2129–2139, DOI: [10.1056/NEJMoa040938](https://doi.org/10.1056/NEJMoa040938).
- 74 P. B. Chapman, *et al.*, Improved survival with vemurafenib in melanoma with BRAF V600E mutation, *N. Engl. J. Med.*, 2011, **364**(26), 2507–2516, DOI: [10.1056/NEJMoa1103782](https://doi.org/10.1056/NEJMoa1103782).
- 75 L. Garcia-Martinez, Y. Zhang, Y. Nakata, H. L. Chan and L. Morey, Epigenetic mechanisms in breast cancer therapy and resistance, *Nat. Commun.*, 2021, **12**(1), 1786, DOI: [10.1038/s41467-021-22024-3](https://doi.org/10.1038/s41467-021-22024-3).
- 76 M. R. Peter, *et al.*, Distinct DNA methylation patterns associated with treatment resistance in metastatic castration resistant prostate cancer, *Sci. Rep.*, 2021, **11**(1), 6630, DOI: [10.1038/s41598-021-85812-3](https://doi.org/10.1038/s41598-021-85812-3).
- 77 E. Leucci, *et al.*, Melanoma addiction to the long non-coding RNA SAMMSON, *Nature*, 2016, **531**(7595), 518–522, DOI: [10.1038/nature17161](https://doi.org/10.1038/nature17161).
- 78 J. Gallon, *et al.*, Chromatin accessibility changes at intergenic regions are associated with ovarian cancer drug resistance, *Clin. Epigenet.*, 2021, **13**(1), 122, DOI: [10.1186/s13148-021-01105-6](https://doi.org/10.1186/s13148-021-01105-6).
- 79 T.-T. Liao and M.-H. Yang, Hybrid Epithelial/Mesenchymal State in Cancer Metastasis: Clinical Significance and Regulatory Mechanisms, *Cells*, 2020, **9**(3), 623, DOI: [10.3390/cells9030623](https://doi.org/10.3390/cells9030623).
- 80 H. Ungefroren, *et al.*, The Quasimesenchymal Pancreatic Ductal Epithelial Cell Line PANC-1-A Useful Model to Study Clonal Heterogeneity and EMT Subtype Shifting, *Cancers*, 2022, **14**(9), 2057, DOI: [10.3390/cancers14092057](https://doi.org/10.3390/cancers14092057).
- 81 B. Dave, V. Mittal, N. M. Tan and J. C. Chang, Epithelial-mesenchymal transition, cancer stem cells and treatment resistance, *Breast Cancer Res.*, 2012, **14**(1), 202, DOI: [10.1186/bcr2938](https://doi.org/10.1186/bcr2938).
- 82 T. Celià-Terrassa and M. K. Jolly, Cancer Stem Cells and Epithelial-to-Mesenchymal Transition in Cancer Metastasis, *Cold Spring Harb. Perspect. Med.*, 2020, **10**(7), a036905, DOI: [10.1101/cshperspect.a036905](https://doi.org/10.1101/cshperspect.a036905).
- 83 S. Christen, *et al.*, Breast Cancer-Derived Lung Metastases Show Increased Pyruvate Carboxylase-Dependent Anaplerosis, *Cell Rep.*, 2016, **17**(3), 837–848, DOI: [10.1016/j.celrep.2016.09.042](https://doi.org/10.1016/j.celrep.2016.09.042).
- 84 C. Zhang, *et al.*, FABP5 promotes lymph node metastasis in cervical cancer by reprogramming fatty acid metabolism, *Theranostics*, 2020, **10**(15), 6561–6580, DOI: [10.7150/thno.44868](https://doi.org/10.7150/thno.44868).
- 85 D. Yang, J. Liu, H. Qian and Q. Zhuang, Cancer-associated fibroblasts: from basic science to anticancer therapy, *Exp. Mol. Med.*, 2023, **55**(7), 1322–1332, DOI: [10.1038/s12276-023-01013-0](https://doi.org/10.1038/s12276-023-01013-0).
- 86 Z. Wang, *et al.*, Metastasis-associated fibroblasts: an emerging target for metastatic cancer, *Biomark. Res.*, 2021, **9**, 47, DOI: [10.1186/s40364-021-00305-9](https://doi.org/10.1186/s40364-021-00305-9).



- 87 B. Chung, *et al.*, Human brain metastatic stroma attracts breast cancer cells via chemokines CXCL16 and CXCL12, *npj Breast Cancer*, 2017, 3, 6, DOI: [10.1038/s41523-017-0008-8](https://doi.org/10.1038/s41523-017-0008-8).
- 88 J. Liu, *et al.*, Association of tumour-associated macrophages with cancer cell EMT, invasion, and metastasis of Kazakh oesophageal squamous cell cancer, *Diagn. Pathol.*, 2019, 14(1), 55, DOI: [10.1186/s13000-019-0834-0](https://doi.org/10.1186/s13000-019-0834-0).
- 89 E. Y. Lin, *et al.*, Macrophages regulate the angiogenic switch in a mouse model of breast cancer, *Cancer Res.*, 2006, 66(23), 11238–11246, DOI: [10.1158/0008-5472.CAN-06-1278](https://doi.org/10.1158/0008-5472.CAN-06-1278).
- 90 E. Y. Lin and J. W. Pollard, Tumor-associated macrophages press the angiogenic switch in breast cancer, *Cancer Res.*, 2007, 67(11), 5064–5066, DOI: [10.1158/0008-5472.CAN-07-0912](https://doi.org/10.1158/0008-5472.CAN-07-0912).
- 91 G. Gunaydin, CAFs Interacting With TAMs in Tumor Microenvironment to Enhance Tumorigenesis and Immune Evasion, *Front. Oncol.*, 2021, 11, 668349, DOI: [10.3389/fonc.2021.668349](https://doi.org/10.3389/fonc.2021.668349).
- 92 M. Raymant, *et al.*, Macrophage-fibroblast JAK/STAT dependent crosstalk promotes liver metastatic outgrowth in pancreatic cancer, *Nat. Commun.*, 2024, 15(1), 3593, DOI: [10.1038/s41467-024-47949-3](https://doi.org/10.1038/s41467-024-47949-3).
- 93 X. Li and J. Wang, Mechanical tumor microenvironment and transduction: cytoskeleton mediates cancer cell invasion and metastasis, *Int. J. Biol. Sci.*, 2020, 16(12), 2014–2028, DOI: [10.7150/ijbs.44943](https://doi.org/10.7150/ijbs.44943).
- 94 Z. Mai, Y. Lin, P. Lin, X. Zhao and L. Cui, Modulating extracellular matrix stiffness: a strategic approach to boost cancer immunotherapy, *Cell Death Dis.*, 2024, 15(5), 307, DOI: [10.1038/s41419-024-06697-4](https://doi.org/10.1038/s41419-024-06697-4).
- 95 R. S. Fischer, *et al.*, Contractility, focal adhesion orientation, and stress fiber orientation drive cancer cell polarity and migration along wavy ECM substrates, *Proc. Natl. Acad. Sci. U. S. A.*, 2021, 118(22), e2021135118, DOI: [10.1073/pnas.2021135118](https://doi.org/10.1073/pnas.2021135118).
- 96 E. Dornier and J. C. Norman, Cancer cells with trapped nuclei cut their way through the extracellular matrix, *Nat. Commun.*, 2018, 9(1), 3954, DOI: [10.1038/s41467-018-06351-6](https://doi.org/10.1038/s41467-018-06351-6).
- 97 C. M. Denais, *et al.*, Nuclear envelope rupture and repair during cancer cell migration, *Science*, 2016, 352(6283), 353–358, DOI: [10.1126/science.aad7297](https://doi.org/10.1126/science.aad7297).
- 98 G. P. de F. Nader, *et al.*, Compromised nuclear envelope integrity drives TREX1-dependent DNA damage and tumor cell invasion, *Cell*, 2021, 184(20), 5230–5246.e22, DOI: [10.1016/j.cell.2021.08.035](https://doi.org/10.1016/j.cell.2021.08.035).
- 99 A. Legátová, M. Pelantová, D. Rösel, J. Brábek and A. Škarková, The emerging role of microtubules in invasion plasticity, *Front. Oncol.*, 2023, 13, 1118171, DOI: [10.3389/fonc.2023.1118171](https://doi.org/10.3389/fonc.2023.1118171).
- 100 L. Winter, C. Abrahamsberg and G. Wiche, Plectin isoform 1b mediates mitochondrion-intermediate filament network linkage and controls organelle shape, *J. Cell Biol.*, 2008, 181(6), 903–911, DOI: [10.1083/jcb.200710151](https://doi.org/10.1083/jcb.200710151).
- 101 R. A. Battaglia, S. Delic, H. Herrmann and N. T. Snider, Vimentin on the move: new developments in cell migration, *F1000Research*, 2018, 7(F1000 Faculty Rev), 1796, DOI: [10.12688/f1000research.15967.1](https://doi.org/10.12688/f1000research.15967.1).
- 102 M. Nagano, D. Hoshino, N. Koshikawa, T. Akizawa and M. Seiki, Turnover of Focal Adhesions and Cancer Cell Migration, *Int. J. Cell Biol.*, 2012, 2012, 310616, DOI: [10.1155/2012/310616](https://doi.org/10.1155/2012/310616).
- 103 C. Ruggiero and E. Lalli, Targeting the cytoskeleton against metastatic dissemination, *Cancer Metastasis Rev.*, 2021, 40(1), 89–140, DOI: [10.1007/s10555-020-09936-0](https://doi.org/10.1007/s10555-020-09936-0).
- 104 X. Xu, *et al.*, Disturbing cytoskeleton by engineered nanomaterials for enhanced cancer therapeutics, *Bioact. Mater.*, 2023, 29, 50–71, DOI: [10.1016/j.bioactmat.2023.06.016](https://doi.org/10.1016/j.bioactmat.2023.06.016).
- 105 Y.-L. Wu, Y.-Y. Hong, H.-L. Zhan, L.-Y. Zhang, S.-G. Wu and F.-X. Zhang, Axillary lymph node removal in de novo metastatic breast cancer, *Gland Surg.*, 2024, 13(7), 1214–1228, DOI: [10.21037/gs-24-130](https://doi.org/10.21037/gs-24-130).
- 106 X. Li, J. Strietz, A. Bleilevens, E. Stickeler and J. Maurer, Chemotherapeutic Stress Influences Epithelial-Mesenchymal Transition and Stemness in Cancer Stem Cells of Triple-Negative Breast Cancer, *Int. J. Mol. Sci.*, 2020, 21(2), 404, DOI: [10.3390/ijms21020404](https://doi.org/10.3390/ijms21020404).
- 107 S. Y. Lee, *et al.*, Induction of metastasis, cancer stem cell phenotype, and oncogenic metabolism in cancer cells by ionizing radiation, *Mol. Cancer*, 2017, 16(1), 10, DOI: [10.1186/s12943-016-0577-4](https://doi.org/10.1186/s12943-016-0577-4).
- 108 H. Zhao, *et al.*, Inflammation and tumor progression: signaling pathways and targeted intervention, *Signal. Transduction Targeted Ther.*, 2021, 6(1), 1–46, DOI: [10.1038/s41392-021-00658-5](https://doi.org/10.1038/s41392-021-00658-5).
- 109 S. I. Grivennikov, F. R. Greten and M. Karin, Immunity, Inflammation, and Cancer, *Cell*, 2010, 140(6), 883–899, DOI: [10.1016/j.cell.2010.01.025](https://doi.org/10.1016/j.cell.2010.01.025).
- 110 G. Bocci, A. Di Paolo and R. Danesi, The pharmacological bases of the antiangiogenic activity of paclitaxel, *Angiogenesis*, 2013, 16(3), 481–492, DOI: [10.1007/s10456-013-9334-0](https://doi.org/10.1007/s10456-013-9334-0).
- 111 G. Ranieri, R. Patruno, E. Ruggieri, S. Montemurro, P. Valerio and D. Ribatti, Vascular endothelial growth factor (VEGF) as a target of bevacizumab in cancer: from the biology to the clinic, *Curr. Med. Chem.*, 2006, 13(16), 1845–1857, DOI: [10.2174/092986706777585059](https://doi.org/10.2174/092986706777585059).
- 112 E. V. Cutsem, *et al.*, Safety and efficacy of first-line bevacizumab with FOLFOX, XELOX, FOLFIRI and fluoropyrimidines in metastatic colorectal cancer: the BEAT study, *Ann. Oncol.*, 2009, 20(11), 1842–1847, DOI: [10.1093/annonc/mdp233](https://doi.org/10.1093/annonc/mdp233).
- 113 M. J. Kwon, Matrix metalloproteinases as therapeutic targets in breast cancer, *Front. Oncol.*, 2023, 12, 1108695, DOI: [10.3389/fonc.2022.1108695](https://doi.org/10.3389/fonc.2022.1108695).
- 114 E. Gobin, *et al.*, A pan-cancer perspective of matrix metalloproteases (MMP) gene expression profile and their diagnostic/prognostic potential, *BMC Cancer*, 2019, 19(1), 581, DOI: [10.1186/s12885-019-5768-0](https://doi.org/10.1186/s12885-019-5768-0).
- 115 L. M. Coussens, B. Fingleton and L. M. Matrisian, Matrix metalloproteinase inhibitors and cancer: trials and



- tribulations, *Science*, 2002, **295**(5564), 2387–2392, DOI: [10.1126/science.1067100](https://doi.org/10.1126/science.1067100).
- 116 R. E. Vandenbroucke and C. Libert, Is there new hope for therapeutic matrix metalloproteinase inhibition?, *Nat. Rev. Drug Discovery*, 2014, **13**(12), 904–927, DOI: [10.1038/nrd4390](https://doi.org/10.1038/nrd4390).
- 117 K. L. Sodek, T. J. Brown and M. J. Ringuette, Collagen I but not Matrigel matrices provide an MMP-dependent barrier to ovarian cancer cell penetration, *BMC Cancer*, 2008, **8**, 223, DOI: [10.1186/1471-2407-8-223](https://doi.org/10.1186/1471-2407-8-223).
- 118 J. Zuo, *et al.*, Overexpression of CXCR4 promotes invasion and migration of non-small cell lung cancer via EGFR and MMP-9, *Oncol. Lett.*, 2017, **14**(6), 7513–7521, DOI: [10.3892/ol.2017.7168](https://doi.org/10.3892/ol.2017.7168).
- 119 X. Sun, *et al.*, CXCL12/CXCR4/CXCR7 Chemokine Axis and Cancer Progression, *Cancer Metastasis Rev.*, 2010, **29**(4), 709–722, DOI: [10.1007/s10555-010-9256-x](https://doi.org/10.1007/s10555-010-9256-x).
- 120 S. Liu, *et al.*, Targeting CXCR4 abrogates resistance to trastuzumab by blocking cell cycle progression and synergizes with docetaxel in breast cancer treatment, *Breast Cancer Res.*, 2023, **25**(1), 62, DOI: [10.1186/s13058-023-01665-w](https://doi.org/10.1186/s13058-023-01665-w).
- 121 D. T. Fearon and T. Janowitz, AMD3100/Plerixafor overcomes immune inhibition by the CXCL12–KRT19 coating on pancreatic and colorectal cancer cells, *Br. J. Cancer*, 2021, **125**(2), 149–151, DOI: [10.1038/s41416-021-01315-y](https://doi.org/10.1038/s41416-021-01315-y).
- 122 B. A. Teicher and S. P. Fricker, CXCL12 (SDF-1)/CXCR4 Pathway in Cancer, *Clin. Cancer Res.*, 2010, **16**(11), 2927–2931, DOI: [10.1158/1078-0432.CCR-09-2329](https://doi.org/10.1158/1078-0432.CCR-09-2329).
- 123 M. D. Galsky, *et al.*, A Phase I Trial of LY2510924, a CXCR4 Peptide Antagonist, in Patients with Advanced Cancer, *Clin. Cancer Res.*, 2014, **20**(13), 3581–3588, DOI: [10.1158/1078-0432.CCR-13-2686](https://doi.org/10.1158/1078-0432.CCR-13-2686).
- 124 B. Bockorny, *et al.*, BL-8040, a CXCR4 antagonist, in combination with pembrolizumab and chemotherapy for pancreatic cancer: the COMBAT trial, *Nat. Med.*, 2020, **26**(6), 878–885, DOI: [10.1038/s41591-020-0880-x](https://doi.org/10.1038/s41591-020-0880-x).
- 125 R. Kumar, *et al.*, Recent developments in receptor tyrosine kinase inhibitors: a promising mainstay in targeted cancer therapy, *Med. Drug Discovery*, 2024, **23**, 100195, DOI: [10.1016/j.medidd.2024.100195](https://doi.org/10.1016/j.medidd.2024.100195).
- 126 S. Faivre, G. Demetri, W. Sargent and E. Raymond, Molecular basis for sunitinib efficacy and future clinical development, *Nat. Rev. Drug Discovery*, 2007, **6**(9), 734–745, DOI: [10.1038/nrd2380](https://doi.org/10.1038/nrd2380).
- 127 C. E. Geyer, *et al.*, Lapatinib plus Capecitabine for HER2-Positive Advanced Breast Cancer, *N. Engl. J. Med.*, 2006, **355**(26), 2733–2743, DOI: [10.1056/NEJMoa064320](https://doi.org/10.1056/NEJMoa064320).
- 128 K.-H. Yeh, *et al.*, Real-world evidence of the safety and effectiveness of regorafenib in Taiwanese patients with metastatic colorectal cancer: CORRELATE Taiwan, *J. Formosan Med. Assoc.*, 2021, **120**(11), 2023–2031, DOI: [10.1016/j.jfma.2020.12.015](https://doi.org/10.1016/j.jfma.2020.12.015).
- 129 L. Meng, *et al.*, Mechanisms of immune checkpoint inhibitors: insights into the regulation of circular RNAs involved in cancer hallmarks, *Cell Death Dis.*, 2024, **15**(1), 1–26, DOI: [10.1038/s41419-023-06389-5](https://doi.org/10.1038/s41419-023-06389-5).
- 130 E. I. Buchbinder and A. Desai, CTLA-4 and PD-1 Pathways, *Am. J. Clin. Oncol.*, 2016, **39**(1), 98–106, DOI: [10.1097/COC.000000000000239](https://doi.org/10.1097/COC.000000000000239).
- 131 S. J. Klemptner, *et al.*, Tumor Mutational Burden as a Predictive Biomarker for Response to Immune Checkpoint Inhibitors: A Review of Current Evidence, *The Oncologist*, 2020, **25**(1), e147–e159, DOI: [10.1634/theoncologist.2019-0244](https://doi.org/10.1634/theoncologist.2019-0244).
- 132 J. H. Park, M. B. Geyer and R. J. Brentjens, CD19-targeted CAR T-cell therapeutics for hematologic malignancies: interpreting clinical outcomes to date, *Blood*, 2016, **127**(26), 3312–3320, DOI: [10.1182/blood-2016-02-629063](https://doi.org/10.1182/blood-2016-02-629063).
- 133 A. Betof Warner, P. G. Corrie and O. Hamid, Tumor-Infiltrating Lymphocyte Therapy in Melanoma: Facts to the Future, *Clin. Cancer Res.*, 2023, **29**(10), 1835–1854, DOI: [10.1158/1078-0432.CCR-22-1922](https://doi.org/10.1158/1078-0432.CCR-22-1922).
- 134 M. J. Besser, *et al.*, Adoptive transfer of tumor-infiltrating lymphocytes in patients with metastatic melanoma: intent-to-treat analysis and efficacy after failure to prior immunotherapies, *Clin. Cancer Res.*, 2013, **19**(17), 4792–4800, DOI: [10.1158/1078-0432.CCR-13-0380](https://doi.org/10.1158/1078-0432.CCR-13-0380).
- 135 A. Gandalovičová, *et al.*, Migrastatics-Anti-metastatic and Anti-invasion Drugs: Promises and Challenges, *Trends Cancer*, 2017, **3**(6), 391–406, DOI: [10.1016/j.trecan.2017.04.008](https://doi.org/10.1016/j.trecan.2017.04.008).
- 136 D. Rosel, M. Fernandes, V. Sanz-Moreno and J. Brábek, Migrastatics: Redirecting R&D in Solid Cancer Towards Metastasis?, *Trends Cancer*, 2019, **5**(12), 755–756, DOI: [10.1016/j.trecan.2019.10.011](https://doi.org/10.1016/j.trecan.2019.10.011).
- 137 J. L. Fialova, *et al.*, Pentamethinium salts suppress key metastatic processes by regulating mitochondrial function and inhibiting dihydroorotate dehydrogenase respiration, *Biomed. Pharmacother.*, 2022, **154**, 113582, DOI: [10.1016/j.biopha.2022.113582](https://doi.org/10.1016/j.biopha.2022.113582).
- 138 W. Tong, Q. Wang, D. Sun and J. Suo, Curcumin suppresses colon cancer cell invasion via AMPK-induced inhibition of NF- κ B, uPA activator and MMP9, *Oncol. Lett.*, 2016, **12**(5), 4139–4146, DOI: [10.3892/ol.2016.5148](https://doi.org/10.3892/ol.2016.5148).
- 139 M. Zahedi, H. Salmani Izadi, F. Arghidash, E. Gumprich, M. Banach and A. Sahebkar, The effect of curcumin on hypoxia in the tumour microenvironment as a regulatory factor in cancer, *Arch. Med. Sci.*, 2023, **19**(6), 1616–1629, DOI: [10.5114/aoms/171122](https://doi.org/10.5114/aoms/171122).
- 140 S. Singh, C. A. Barnes, J. S. D'Souza, R. V. Hosur and P. Mishra, Curcumin, a potential initiator of apoptosis via direct interactions with Bcl-xL and Bid, *Proteins Struct., Funct., Bioinf.*, 2022, **90**(2), 455–464, DOI: [10.1002/prot.26238](https://doi.org/10.1002/prot.26238).
- 141 V. P. Kale, *et al.*, A novel selective multikinase inhibitor of ROCK and MRCK effectively blocks cancer cell migration and invasion, *Cancer Lett.*, 2014, **354**(2), 299–310, DOI: [10.1016/j.canlet.2014.08.032](https://doi.org/10.1016/j.canlet.2014.08.032).
- 142 M. Raudenská, *et al.*, Engine shutdown: migrastatic strategies and prevention of metastases, *Trends Cancer*, 2023, **9**(4), 293–308, DOI: [10.1016/j.trecan.2023.01.001](https://doi.org/10.1016/j.trecan.2023.01.001).
- 143 J. Conde, N. Oliva and N. Artzi, Implantable hydrogel embedded dark-gold nanoswitch as a theranostic probe



- to sense and overcome cancer multidrug resistance, *Proc. Natl. Acad. Sci. U. S. A.*, 2015, **112**(11), E1278–E1287, DOI: [10.1073/pnas.1421229112](https://doi.org/10.1073/pnas.1421229112).
- 144 P. Calvo, *et al.*, Quantification and localization of PEGylated polycyanoacrylate nanoparticles in brain and spinal cord during experimental allergic encephalomyelitis in the rat, *Eur. J. Neurosci.*, 2002, **15**(8), 1317–1326, DOI: [10.1046/j.1460-9568.2002.01967.x](https://doi.org/10.1046/j.1460-9568.2002.01967.x).
- 145 Y.-Q. Wang, *et al.*, Prolonged blood circulation outperforms active targeting for nanocarriers-mediated enhanced hepatocellular carcinoma therapy in vivo, *J. Controlled Release*, 2022, **347**, 400–413, DOI: [10.1016/j.jconrel.2022.05.024](https://doi.org/10.1016/j.jconrel.2022.05.024).
- 146 T. Paresishvili and Z. Kakabadze, Challenges and Opportunities Associated With Drug Delivery for the Treatment of Solid Tumors, *Oncol. Rev.*, 2023, **17**, 10577, DOI: [10.3389/or.2023.10577](https://doi.org/10.3389/or.2023.10577).
- 147 J. Wu, The Enhanced Permeability and Retention (EPR) Effect: The Significance of the Concept and Methods to Enhance Its Application, *J. Pers. Med.*, 2021, **11**(8), 771, DOI: [10.3390/jpm11080771](https://doi.org/10.3390/jpm11080771).
- 148 H. Maeda, Toward a full understanding of the EPR effect in primary and metastatic tumors as well as issues related to its heterogeneity, *Adv. Drug Delivery Rev.*, 2015, **91**, 3–6, DOI: [10.1016/j.addr.2015.01.002](https://doi.org/10.1016/j.addr.2015.01.002).
- 149 D. Rosenblum, N. Joshi, W. Tao, J. M. Karp and D. Peer, Progress and challenges towards targeted delivery of cancer therapeutics, *Nat. Commun.*, 2018, **9**(1), 1410, DOI: [10.1038/s41467-018-03705-y](https://doi.org/10.1038/s41467-018-03705-y).
- 150 P. S. Gill, *et al.*, Randomized phase III trial of liposomal daunorubicin versus doxorubicin, bleomycin, and vincristine in AIDS-related Kaposi's sarcoma, *J. Clin. Oncol.*, 1996, **14**(8), 2353–2364, DOI: [10.1200/JCO.1996.14.8.2353](https://doi.org/10.1200/JCO.1996.14.8.2353).
- 151 D. D. V. Hoff, *et al.*, Increased Survival in Pancreatic Cancer with nab-Paclitaxel plus Gemcitabine, *N. Engl. J. Med.*, 2013, **369**(18), 1691–1703, DOI: [10.1056/NEJMoa1304369](https://doi.org/10.1056/NEJMoa1304369).
- 152 M. Sousa de Almeida, E. Susnik, B. Drasler, P. Taladriz-Blanco, A. Petri-Fink and B. Rothen-Rutishauser, Understanding nanoparticle endocytosis to improve targeting strategies in nanomedicine, *Chem. Soc. Rev.*, 2021, **50**(9), 5397–5434, DOI: [10.1039/d0cs01127d](https://doi.org/10.1039/d0cs01127d).
- 153 M. Sanati, A. R. Afshari, S. Aminyavari, P. Kesharwani, T. Jamialahmadi and A. Sahebkar, RGD-engineered nanoparticles as an innovative drug delivery system in cancer therapy, *J. Drug Delivery Sci. Technol.*, 2023, **84**, 104562, DOI: [10.1016/j.jddst.2023.104562](https://doi.org/10.1016/j.jddst.2023.104562).
- 154 E. Ruoslahti, RGD and other recognition sequences for integrins, *Annu. Rev. Cell Dev. Biol.*, 1996, **12**, 697–715, DOI: [10.1146/annurev.cellbio.12.1.697](https://doi.org/10.1146/annurev.cellbio.12.1.697).
- 155 H. Javid, M. A. Oryani, N. Rezagholinejad, A. Esparham, M. Tajaldini and M. Karimi-Shahri, RGD peptide in cancer targeting: benefits, challenges, solutions, and possible integrin–RGD interactions, *Cancer Med.*, 2024, **13**(2), e6800, DOI: [10.1002/cam4.6800](https://doi.org/10.1002/cam4.6800).
- 156 S. M. Weis and D. A. Cheresh, α v Integrins in Angiogenesis and Cancer, *Cold Spring Harb. Perspect. Med.*, 2011, **1**(1), a006478, DOI: [10.1101/cshperspect.a006478](https://doi.org/10.1101/cshperspect.a006478).
- 157 T. R. Daniels, *et al.*, Transferrin receptors and the targeted delivery of therapeutic agents against cancer, *Biochim. Biophys. Acta*, 2012, **1820**(3), 291–317, DOI: [10.1016/j.bbagen.2011.07.016](https://doi.org/10.1016/j.bbagen.2011.07.016).
- 158 A. K. Grewal and R. K. Salar, Chitosan nanoparticle delivery systems: an effective approach to enhancing efficacy and safety of anticancer drugs, *Nano TransMed*, 2024, **3**, 100040, DOI: [10.1016/j.ntm.2024.100040](https://doi.org/10.1016/j.ntm.2024.100040).
- 159 A. R. Jordan, R. R. Racine, M. J. P. Hennig and V. B. Lokeshwar, The Role of CD44 in Disease Pathophysiology and Targeted Treatment, *Front. Immunol.*, 2015, **6**, 182, DOI: [10.3389/fimmu.2015.00182](https://doi.org/10.3389/fimmu.2015.00182).
- 160 P. Kesharwani, R. Chadar, A. Sheikh, W. Y. Rizg and A. Y. Safhi, CD44-Targeted Nanocarrier for Cancer Therapy, *Front. Pharmacol.*, 2022, **12**, 800481, DOI: [10.3389/fphar.2021.800481](https://doi.org/10.3389/fphar.2021.800481).
- 161 T. Ooki, N. Murata-Kamiya, A. Takahashi-Kanemitsu, W. Wu and M. Hatakeyama, High-Molecular-Weight Hyaluronan Is a Hippo Pathway Ligand Directing Cell Density-Dependent Growth Inhibition via PAR1b, *Dev. Cell*, 2019, **49**(4), 590–604.e9, DOI: [10.1016/j.devcel.2019.04.018](https://doi.org/10.1016/j.devcel.2019.04.018).
- 162 A. G. Tavianatou, I. Caon, M. Franchi, Z. Piperigkou, D. Galesso and N. K. Karamanos, Hyaluronan: molecular size-dependent signaling and biological functions in inflammation and cancer, *FEBS J.*, 2019, **286**(15), 2883–2908, DOI: [10.1111/febs.14777](https://doi.org/10.1111/febs.14777).
- 163 G. Nabil, *et al.*, CD44 Targeted Nanomaterials for Treatment of Triple-Negative Breast Cancer, *Cancers*, 2021, **13**(4), 898, DOI: [10.3390/cancers13040898](https://doi.org/10.3390/cancers13040898).
- 164 Y. Barenholz, Doxil[®]—the first FDA-approved nano-drug: lessons learned, *J. Controlled Release*, 2012, **160**(2), 117–134, DOI: [10.1016/j.jconrel.2012.03.020](https://doi.org/10.1016/j.jconrel.2012.03.020).
- 165 A. Moreno-Aspitia and E. A. Perez, Nanoparticle albumin-bound paclitaxel (ABI-007): a newer taxane alternative in breast cancer, *Future Oncol. Lond. Engl.*, 2005, **1**(6), 755–762, DOI: [10.2217/14796694.1.6.755](https://doi.org/10.2217/14796694.1.6.755).
- 166 Y. Cheng, *et al.*, Nanoparticulates reduce tumor cell migration through affinity interactions with extracellular migrasomes and retraction fibers, *Nanoscale Horiz.*, 2022, **7**(7), 779–789, DOI: [10.1039/d2nh00067a](https://doi.org/10.1039/d2nh00067a).
- 167 L.-W. Tong, *et al.*, Synergistic anti-tumor effect of dual drug co-assembled nanoparticles based on ursolic acid and sorafenib, *Colloids Surf., B*, 2024, **234**, 113724, DOI: [10.1016/j.colsurfb.2023.113724](https://doi.org/10.1016/j.colsurfb.2023.113724).
- 168 A. A. H. Abdellatif, A. T. Ali, A. Bouazzaoui, M. Alsharidah, O. A. Rugaie and N. S. Tolba, Formulation of polymeric nanoparticles loaded sorafenib; evaluation of cytotoxicity, molecular evaluation, and gene expression studies in lung and breast cancer cell lines, *Nanotechnol. Rev.*, 2022, **11**(1), 987–1004, DOI: [10.1515/ntrev-2022-0058](https://doi.org/10.1515/ntrev-2022-0058).
- 169 I. Fatima, *et al.*, Advances in targeted nano-delivery of bevacizumab using nanoparticles: current insights, innovations, and future perspectives, *J. Drug Delivery Sci. Technol.*, 2024, **98**, 105850, DOI: [10.1016/j.jddst.2024.105850](https://doi.org/10.1016/j.jddst.2024.105850).
- 170 F. Sousa, H. K. Dhaliwal, F. Gattacceca, B. Sarmiento and M. M. Amiji, Enhanced anti-angiogenic effects of



- bevacizumab in glioblastoma treatment upon intranasal administration in polymeric nanoparticles, *J. Controlled Release*, 2019, **309**, 37–47, DOI: [10.1016/j.jconrel.2019.07.033](https://doi.org/10.1016/j.jconrel.2019.07.033).
- 171 M. M. Ahmed, *et al.*, Boosting the Anticancer Activity of Sunitinib Malate in Breast Cancer through Lipid Polymer Hybrid Nanoparticles Approach, *Polymers*, 2022, **14**(12), 2459, DOI: [10.3390/polym14122459](https://doi.org/10.3390/polym14122459).
- 172 Z.-J. Huo, *et al.*, Novel nanosystem to enhance the anti-tumor activity of lapatinib in breast cancer treatment: therapeutic efficacy evaluation, *Cancer Sci.*, 2015, **106**(10), 1429–1437, DOI: [10.1111/cas.12737](https://doi.org/10.1111/cas.12737).
- 173 X. Wu, *et al.*, Co-loaded lapatinib/PAB by ferritin nanoparticles eliminated ECM-detached cluster cells via modulating EGFR in triple-negative breast cancer, *Cell Death Dis.*, 2022, **13**(6), 1–13, DOI: [10.1038/s41419-022-05007-0](https://doi.org/10.1038/s41419-022-05007-0).
- 174 Z. Yao, *et al.*, Hollow Cu₂MoS₄ nanoparticles loaded with immune checkpoint inhibitors reshape the tumor micro-environment to enhance immunotherapy for pancreatic cancer, *Acta Biomater.*, 2024, **173**, 365–377, DOI: [10.1016/j.actbio.2023.10.024](https://doi.org/10.1016/j.actbio.2023.10.024).
- 175 A. Nayak, S. Das, D. Nayak, C. Sethy, S. Narayan and C. N. Kundu, Nanoquinacrine sensitizes 5-FU-resistant cervical cancer stem-like cells by down-regulating Nectin-4 via ADAM-17 mediated NOTCH deregulation, *Cell. Oncol.*, 2019, **42**(2), 157–171, DOI: [10.1007/s13402-018-0417-1](https://doi.org/10.1007/s13402-018-0417-1).
- 176 L. Luo, *et al.*, Stimuli-responsive polymeric prodrug-based nanomedicine delivering nifuroxazide and doxorubicin against primary breast cancer and pulmonary metastasis, *J. Controlled Release*, 2020, **318**, 124–135, DOI: [10.1016/j.jconrel.2019.12.017](https://doi.org/10.1016/j.jconrel.2019.12.017).
- 177 R. Guo, *et al.*, Development of a Non-Coding-RNA-based EMT/CSC Inhibitory Nanomedicine for In Vivo Treatment and Monitoring of HCC, *Adv. Sci.*, 2019, **6**(9), 1801885, DOI: [10.1002/advs.201801885](https://doi.org/10.1002/advs.201801885).
- 178 F. Ding, L. Zhang, H. Chen, H. Song, S. Chen and H. Xiao, Enhancing the chemotherapeutic efficacy of platinum prodrug nanoparticles and inhibiting cancer metastasis by targeting iron homeostasis, *Nanoscale Horiz.*, 2020, **5**(6), 999–1015, DOI: [10.1039/d0nh00148a](https://doi.org/10.1039/d0nh00148a).
- 179 Y. Zhu, *et al.*, Enhanced Anti-Metastatic Activity of Etoposide Using Layered Double Hydroxide Nano Particles, *J. Biomed. Nanotechnol.*, 2015, **11**(12), 2158–2168, DOI: [10.1166/jbn.2015.2164](https://doi.org/10.1166/jbn.2015.2164).
- 180 L. Zhao, C. Gu, Y. Gan, L. Shao, H. Chen and H. Zhu, Exosome-mediated siRNA delivery to suppress postoperative breast cancer metastasis, *J. Controlled Release*, 2020, **318**, 1–15, DOI: [10.1016/j.jconrel.2019.12.005](https://doi.org/10.1016/j.jconrel.2019.12.005).
- 181 X. Zhang, *et al.*, Bone-targeted nanoplatform enables efficient modulation of bone tumor microenvironment for prostate cancer bone metastasis treatment, *Drug Delivery*, 2022, **29**(1), 889–905, DOI: [10.1080/10717544.2022.2050845](https://doi.org/10.1080/10717544.2022.2050845).
- 182 W. Sun, *et al.*, Bone-Targeted Nanoplatform Combining Zoledronate and Photothermal Therapy To Treat Breast Cancer Bone Metastasis, *ACS Nano*, 2019, **13**(7), 7556–7567, DOI: [10.1021/acsnano.9b00097](https://doi.org/10.1021/acsnano.9b00097).
- 183 Y. Zhou, *et al.*, Targeted Delivery of Secretory Promelittin via Novel Poly(lactone-co-β-amino ester) Nanoparticles for Treatment of Breast Cancer Brain Metastases, *Adv. Sci.*, 2020, **7**(5), 1901866, DOI: [10.1002/advs.201901866](https://doi.org/10.1002/advs.201901866).
- 184 L. Kaps and D. Schuppan, Targeting Cancer Associated Fibroblasts in Liver Fibrosis and Liver Cancer Using Nanocarriers, *Cells*, 2020, **9**(9), 9, DOI: [10.3390/cells9092027](https://doi.org/10.3390/cells9092027).
- 185 M. H. M. G. M. den Brok, *et al.*, In situ tumor ablation creates an antigen source for the generation of antitumor immunity, *Cancer Res.*, 2004, **64**(11), 4024–4029, DOI: [10.1158/0008-5472.CAN-03-3949](https://doi.org/10.1158/0008-5472.CAN-03-3949).
- 186 S. P. Haen, P. L. Pereira, H. R. Salih, H.-G. Rammensee and C. Gouttefangeas, More Than Just Tumor Destruction: Immunomodulation by Thermal Ablation of Cancer, *Clin. Dev. Immunol.*, 2011, **2011**, 160250, DOI: [10.1155/2011/160250](https://doi.org/10.1155/2011/160250).
- 187 A. Perez-Potti, M. Rodríguez-Pérez, E. Polo, B. Pelaz and P. del Pino, Nanoparticle-based immunotherapeutics: from the properties of nanocores to the differential effects of administration routes, *Adv. Drug Delivery Rev.*, 2023, **197**, 114829, DOI: [10.1016/j.addr.2023.114829](https://doi.org/10.1016/j.addr.2023.114829).
- 188 X. Deng, Z. Shao and Y. Zhao, Solutions to the Drawbacks of Photothermal and Photodynamic Cancer Therapy, *Adv. Sci.*, 2021, **8**(3), 2002504, DOI: [10.1002/advs.202002504](https://doi.org/10.1002/advs.202002504).
- 189 Y. Ding, *et al.*, NO-releasing polypeptide nanocomposites reverse cancer multidrug resistance via triple therapies, *Acta Biomater.*, 2021, **123**, 335–345, DOI: [10.1016/j.actbio.2021.01.015](https://doi.org/10.1016/j.actbio.2021.01.015).
- 190 Y. Tian, Y. Lei, Y. Wang, J. Lai, J. Wang and F. Xia, Mechanism of multidrug resistance to chemotherapy mediated by P-glycoprotein (Review), *Int. J. Oncol.*, 2023, **63**(5), 119, DOI: [10.3892/ijo.2023.5567](https://doi.org/10.3892/ijo.2023.5567).
- 191 Y. Li, *et al.*, Multipronged Design of Light-Triggered Nanoparticles To Overcome Cisplatin Resistance for Efficient Ablation of Resistant Tumor, *ACS Nano*, 2015, **9**(10), 9626–9637, DOI: [10.1021/acsnano.5b05097](https://doi.org/10.1021/acsnano.5b05097).
- 192 A. A. A. Al-Ali, N. Al Ward, M. A. Obeid, C. Uhd Nielsen, P. A. Mulheran and M. M. Al Qaraghuli, Recent Advances in Photothermal Therapies Against Cancer and the Role of Membrane Transporter Modulators on the Efficacy of This Approach, *Technol. Cancer Res. Treat.*, 2023, **22**, 15330338231168016, DOI: [10.1177/15330338231168016](https://doi.org/10.1177/15330338231168016).
- 193 R. S. Riley and E. S. Day, Gold nanoparticle-mediated photothermal therapy: applications and opportunities for multimodal cancer treatment, *Wiley Interdiscip. Rev.: Nanomed. Nanobiotechnol.*, 2017, **9**(4), e1449, DOI: [10.1002/wnan.1449](https://doi.org/10.1002/wnan.1449).
- 194 J. Ma, N. Li, J. Wang, Z. Liu, Y. Han and Y. Zeng, In vivo synergistic tumor therapies based on copper sulfide photothermal therapeutic nanoplatforms, *Exploration*, 2023, **3**(5), 20220161, DOI: [10.1002/EXP.20220161](https://doi.org/10.1002/EXP.20220161).
- 195 Y. Qiu, *et al.*, Nano Ultrasound Contrast Agent for Synergistic Chemo-photothermal Therapy and Enhanced Immunotherapy Against Liver Cancer and Metastasis, *Adv. Sci.*, 2023, **10**(21), 2300878, DOI: [10.1002/advs.202300878](https://doi.org/10.1002/advs.202300878).
- 196 S. Kim, *et al.*, In situ immunogenic clearance induced by a combination of photodynamic therapy and rho-kinase



- inhibition sensitizes immune checkpoint blockade response to elicit systemic antitumor immunity against intraocular melanoma and its metastasis, *J. Immunother. Cancer*, 2021, **9**(1), e001481, DOI: [10.1136/jitc-2020-001481](https://doi.org/10.1136/jitc-2020-001481).
- 197 M. T. Luiz, *et al.*, Targeted Liposomes: A Nonviral Gene Delivery System for Cancer Therapy, *Pharmaceutics*, 2022, **14**(4), 821, DOI: [10.3390/pharmaceutics14040821](https://doi.org/10.3390/pharmaceutics14040821).
- 198 S. A. Shahin, *et al.*, Hyaluronic acid conjugated nanoparticle delivery of siRNA against TWIST reduces tumor burden and enhances sensitivity to cisplatin in ovarian cancer, *Nanomed. Nanotechnol. Biol. Med.*, 2018, **14**(4), 1381–1394, DOI: [10.1016/j.nano.2018.04.008](https://doi.org/10.1016/j.nano.2018.04.008).
- 199 D. Chen, *et al.*, Selective mediation of ovarian cancer SKOV3 cells death by pristine carbon quantum dots/Cu2O composite through targeting matrix metalloproteinases, angiogenic cytokines and cytoskeleton, *J. Nanobiotechnol.*, 2021, **19**(1), 68, DOI: [10.1186/s12951-021-00813-8](https://doi.org/10.1186/s12951-021-00813-8).
- 200 V. M. Díaz-García, *et al.*, Biomimetic quantum dot-labeled B16F10 murine melanoma cells as a tool to monitor early steps of lung metastasis by in vivo imaging, *Int. J. Nanomed.*, 2018, **13**, 6391–6412, DOI: [10.2147/IJN.S165565](https://doi.org/10.2147/IJN.S165565).
- 201 A. Roshini, *et al.*, pH-sensitive tangeretin-ZnO quantum dots exert apoptotic and anti-metastatic effects in metastatic lung cancer cell line, *Mater. Sci. Eng., C*, 2018, **92**, 477–488, DOI: [10.1016/j.msec.2018.06.073](https://doi.org/10.1016/j.msec.2018.06.073).
- 202 P. Ih, *et al.*, An Open-Label, Randomized, Parallel, Phase III Trial Evaluating the Efficacy and Safety of Polymeric Micelle-Formulated Paclitaxel Compared to Conventional Cremophor EL-Based Paclitaxel for Recurrent or Metastatic HER2-Negative Breast Cancer, *Cancer Res. Treat.*, 2017, **49**(3), 569–577, DOI: [10.4143/crt.2016.289](https://doi.org/10.4143/crt.2016.289).
- 203 D. Chen, B. Li, T. Lei, D. Na, M. Nie, Y. Yang, C. Xie, Z. He and J. Wang, Selective mediation of ovarian cancer SKOV3 cells death by pristine carbon quantum dots/Cu2O composite through targeting matrix metalloproteinases, angiogenic cytokines and cytoskeleton, *J. Nanobiotechnol.*, 2021, **19**(1), 68, DOI: [10.1186/s12951-021-00813-8](https://doi.org/10.1186/s12951-021-00813-8).
- 204 C. F. Jehn, P. Hemmati, S. Lehenbauer-Dehm, S. Kümmel, B. Flath and P. Schmid, Biweekly Pegylated Liposomal Doxorubicin (Caelyx) in Heavily Pretreated Metastatic Breast Cancer: A Phase 2 Study, *Clin. Breast Cancer*, 2016, **16**(6), 514–519, DOI: [10.1016/j.clbc.2016.06.001](https://doi.org/10.1016/j.clbc.2016.06.001).
- 205 M. Salzberg, *et al.*, Pegylated liposomal doxorubicin (caelyx) in metastatic breast cancer: a community-based observation study, *Oncology*, 2007, **72**(3–4), 147–151, DOI: [10.1159/000112731](https://doi.org/10.1159/000112731).
- 206 Caelyx pegylated liposomal | European Medicines Agency (EMA), Accessed: Mar. 30, 2026. [Online]. Available: <https://www.ema.europa.eu/en/medicines/human/EPAR/caelyx-pegylated-liposomal>.
- 207 V. Lorusso, *et al.*, Use of Liposomal Doxorubicin for Metastatic Breast Cancer Management Across Europe: Results of Eos (European Observatory & Survey), *Ann. Oncol.*, 2014, **25**, iv130, DOI: [10.1093/annonc/mdu329.45](https://doi.org/10.1093/annonc/mdu329.45).
- 208 Myocet liposomal (previously Myocet) | European Medicines Agency (EMA), Accessed: Mar. 30, 2026. [Online]. Available: <https://www.ema.europa.eu/en/medicines/human/EPAR/myocet-liposomal>.
- 209 X. Xu, L. Wang, H.-Q. Xu, X.-E. Huang, Y.-D. Qian and J. Xiang, Clinical comparison between paclitaxel liposome (Lipusu[®]) and paclitaxel for treatment of patients with metastatic gastric cancer, *Asian Pac. J. Cancer Prev.*, 2013, **14**(4), 2591–2594, DOI: [10.7314/apjcp.2013.14.4.2591](https://doi.org/10.7314/apjcp.2013.14.4.2591).
- 210 J. Y. Jung, *et al.*, A Phase II Study of Genexol(R) (paclitaxel) in Metastatic Breast Cancer, *Cancer Res. Treat.*, 2001, **33**(6), 451–457, DOI: [10.4143/crt.2001.33.6.451](https://doi.org/10.4143/crt.2001.33.6.451).
- 211 K. S. Lee, *et al.*, Multicenter phase II trial of Genexol-PM, a Cremophor-free, polymeric micelle formulation of paclitaxel, in patients with metastatic breast cancer, *Breast Cancer Res. Treat.*, 2008, **108**(2), 241–250, DOI: [10.1007/s10549-007-9591-y](https://doi.org/10.1007/s10549-007-9591-y).
- 212 Z. A. Wainberg, *et al.*, NALIRIFOX versus nab-paclitaxel and gemcitabine in treatment-naive patients with metastatic pancreatic ductal adenocarcinoma (NAPOLI 3): a randomised, open-label, phase 3 trial, *Lancet*, 2023, **402**(10409), 1272–1281, DOI: [10.1016/S0140-6736\(23\)01366-1](https://doi.org/10.1016/S0140-6736(23)01366-1).
- 213 Onivyde pegylated liposomal (previously Onivyde) | European Medicines Agency (EMA), Accessed: Mar. 30, 2026. [Online]. Available: <https://www.ema.europa.eu/en/medicines/human/EPAR/onivyde-pegylated-liposomal>.
- 214 Pazehir | European Medicines Agency (EMA), Accessed: Mar. 25, 2026. [Online]. Available: <https://www.ema.europa.eu/en/medicines/human/EPAR/pazehir>.
- 215 Paclitaxel (Pazehir) 5 mg/ml Powder for Dispersion for Infusion – Summary of Product Characteristics (SmPC) – (emc) | 12063, Accessed: Mar. 30, 2026. [Online]. Available: <https://www.medicines.org.uk/emc/product/12063/smpc>.
- 216 Abraxane | European Medicines Agency (EMA), Accessed: Mar. 25, 2026. [Online]. Available: <https://www.ema.europa.eu/en/medicines/human/EPAR/abraxane>.
- 217 Search Orphan Drug Designations and Approvals, Accessed: Mar. 30, 2026. [Online]. Available: <https://www.accessdata.fda.gov/scripts/opdlisting/opd/detailedIndex.cfm?cfgridkey=73693>.
- 218 DepoCytte | European Medicines Agency (EMA), Accessed: Mar. 30, 2026. [Online]. Available: <https://www.ema.europa.eu/en/medicines/human/EPAR/depocytte>.
- 219 S. Pandit, D. Dutta and S. Nie, Active transcytosis and new opportunities for cancer nanomedicine, *Nat. Mater.*, 2020, **19**(5), 478–480, DOI: [10.1038/s41563-020-0672-1](https://doi.org/10.1038/s41563-020-0672-1).
- 220 S. Chen, *et al.*, Enhanced tumour penetration and prolonged circulation in blood of polyzwitterion–drug conjugates with cell-membrane affinity, *Nat. Biomed. Eng.*, 2021, **5**(9), 1019–1037, DOI: [10.1038/s41551-021-00701-4](https://doi.org/10.1038/s41551-021-00701-4).
- 221 Y. Liu, *et al.*, Transcytosis of Nanomedicine for Tumor Penetration, *Nano Lett.*, 2019, **19**(11), 8010–8020, DOI: [10.1021/acs.nanolett.9b03211](https://doi.org/10.1021/acs.nanolett.9b03211).
- 222 Q. Zhou, *et al.*, Enzyme-activatable polymer–drug conjugate augments tumour penetration and treatment efficacy, *Nat. Nanotechnol.*, 2019, **14**(8), 799–809, DOI: [10.1038/s41565-019-0485-z](https://doi.org/10.1038/s41565-019-0485-z).



- 223 S. Zhang, *et al.*, Transcytosis-Triggering Nanoparticles for Overcoming Stromal Barriers and Reversing Immunosuppression in Pancreatic Cancer Combinatorial Therapy, *Nano Lett.*, 2025, 25(7), 2949–2959, DOI: [10.1021/acs.nanolett.4c06372](https://doi.org/10.1021/acs.nanolett.4c06372).
- 224 C. Caro, J. M. Paez-Muñoz, M. Pernía Leal, M. Carayol, M. Feijoo-Cuaresma and M. L. García-Martín, Metabolically-Driven Active Targeting of Magnetic Nanoparticles Functionalized with Glucuronic Acid to Glioblastoma: Application to MRI-Tracked Magnetic Hyperthermia Therapy, *Adv. Healthcare Mater.*, 2025, 14(3), 2404391, DOI: [10.1002/adhm.202404391](https://doi.org/10.1002/adhm.202404391).
- 225 R. Cheng and S. Wang, Cell-mediated nanoparticle delivery systems: towards precision nanomedicine, *Drug Delivery Transl. Res.*, 2024, 14(11), 3032–3054, DOI: [10.1007/s13346-024-01591-0](https://doi.org/10.1007/s13346-024-01591-0).
- 226 P. Zhang, D. Chen, L. Li and K. Sun, Charge reversal nanosystems for tumor therapy, *J. Nanobiotechnol.*, 2022, 20(1), 31, DOI: [10.1186/s12951-021-01221-8](https://doi.org/10.1186/s12951-021-01221-8).
- 227 H.-R. Moon, B. K. Cho, S. H. Kang, J. H. Ryu and I. C. Kwon, Targeting and exploiting macropinocytosis in cancer therapy, *J. Controlled Release*, 2025, 385, 113962, DOI: [10.1016/j.jconrel.2025.113962](https://doi.org/10.1016/j.jconrel.2025.113962).
- 228 R. R. Arvizo, S. Saha, E. Wang, J. D. Robertson, R. Bhattacharya and P. Mukherjee, Inhibition of tumor growth and metastasis by a self-therapeutic nanoparticle, *Proc. Natl. Acad. Sci. U. S. A.*, 2013, 110(17), 6700–6705, DOI: [10.1073/pnas.1214547110](https://doi.org/10.1073/pnas.1214547110).
- 229 A. Zamborlin, *et al.*, Drug-Free Hybrid Nanoarchitecture Modulation of the Metastatic Behavior of Pancreatic Ductal Adenocarcinoma in Alternative in Vivo Models, *ACS Appl. Nano Mater.*, 2023, 6(24), 22532–22544, DOI: [10.1021/acsanm.3c05299](https://doi.org/10.1021/acsanm.3c05299).
- 230 S. Shao, *et al.*, A non-cytotoxic dendrimer with innate and potent anticancer and anti-metastatic activities, *Nat. Biomed. Eng.*, 2017, 1(9), 745–757, DOI: [10.1038/s41551-017-0130-9](https://doi.org/10.1038/s41551-017-0130-9).
- 231 Y.-T. Lee, *et al.*, Drug-Free Mesoporous Silica Nanoparticles Enable Suppression of Cancer Metastasis and Confer Survival Advantages to Mice with Tumor Xenografts, *ACS Appl. Mater. Interfaces*, 2024, 16(45), 61787–61804, DOI: [10.1021/acsami.4c16609](https://doi.org/10.1021/acsami.4c16609).
- 232 L. García-Hevia, *et al.*, Carbon nanotubes targeted to the tumor microenvironment inhibit metastasis in a preclinical model of melanoma, *Bioact. Mater.*, 2024, 34, 237–247, DOI: [10.1016/j.bioactmat.2023.12.013](https://doi.org/10.1016/j.bioactmat.2023.12.013).
- 233 K. Brzóska, *et al.*, Silver Nanoparticles Inhibit Metastasis of 4T1 Tumor in Mice after Intragastric but Not Intravenous Administration, *Materials*, 2022, 15(11), 3837, DOI: [10.3390/ma15113837](https://doi.org/10.3390/ma15113837).
- 234 S. Sarkar, *et al.*, In situ incorporation of boronate into carbonized alginate nanogels for targeted inhibition of triple-negative breast cancer metastasis by inducing cytoskeletal disruption, cell growth arrest, and apoptosis, *Biomaterials*, 2026, 324, 123500, DOI: [10.1016/j.biomaterials.2025.123500](https://doi.org/10.1016/j.biomaterials.2025.123500).
- 235 J. Hu, *et al.*, In Situ Generated and Amplified Oxidative Stress with Metallo-Nanodrug Assembly for Metastatic Cancer Therapy with High Specificity and Efficacy, *Adv. Ther.*, 2021, 4(11), 2100148, DOI: [10.1002/adtp.202100148](https://doi.org/10.1002/adtp.202100148).
- 236 S. K. Golombek, *et al.*, Tumor targeting via EPR: strategies to enhance patient responses, *Adv. Drug Delivery Rev.*, 2018, 130, 17–38, DOI: [10.1016/j.addr.2018.07.007](https://doi.org/10.1016/j.addr.2018.07.007).
- 237 S. Wilhelm, *et al.*, Analysis of nanoparticle delivery to tumours, *Nat. Rev. Mater.*, 2016, 1(5), 16014, DOI: [10.1038/natrevmats.2016.14](https://doi.org/10.1038/natrevmats.2016.14).
- 238 H. Maeda, Toward a full understanding of the EPR effect in primary and metastatic tumors as well as issues related to its heterogeneity, *Adv. Drug Delivery Rev.*, 2015, 91, 3–6, DOI: [10.1016/j.addr.2015.01.002](https://doi.org/10.1016/j.addr.2015.01.002).
- 239 M. A. Miller, *et al.*, Predicting therapeutic nanoparticle efficacy using a companion MR imaging nanoparticle, *Sci. Transl. Med.*, 2015, 7(314), 314ra183, DOI: [10.1126/scitranslmed.aac6522](https://doi.org/10.1126/scitranslmed.aac6522).
- 240 P. C. Ke, S. Lin, W. J. Parak, T. P. Davis and F. Caruso, A Decade of the Protein Corona, *ACS Nano*, 2017, 11(12), 11773–11776, DOI: [10.1021/acs.nano.7b08008](https://doi.org/10.1021/acs.nano.7b08008).
- 241 D. Docter, D. Westmeier, M. Markiewicz, S. Stolte, S. K. Knauer and R. H. Stauber, The nanoparticle biomolecule corona: lessons learned – challenge accepted?, *Chem. Soc. Rev.*, 2015, 44(17), 6094–6121, DOI: [10.1039/c5cs00217f](https://doi.org/10.1039/c5cs00217f).
- 242 Y. Boucher, L. T. Baxter and R. K. Jain, Interstitial pressure gradients in tissue-isolated and subcutaneous tumors: implications for therapy, *Cancer Res.*, 1990, 50(15), 4478–4484.
- 243 S. K. Balasubramanian, J. Jittiwat, J. Manikandan, C.-N. Ong, L. E. Yu and W.-Y. Ong, Biodistribution of gold nanoparticles and gene expression changes in the liver and spleen after intravenous administration in rats, *Biomaterials*, 2010, 31(8), 2034–2042, DOI: [10.1016/j.biomaterials.2009.11.079](https://doi.org/10.1016/j.biomaterials.2009.11.079).
- 244 R. S. H. Yang, *et al.*, Persistent Tissue Kinetics and Redistribution of Nanoparticles, Quantum Dot 705, in Mice: ICP-MS Quantitative Assessment, *Environ. Health Perspect.*, 2007, 115(9), 1339–1343, DOI: [10.1289/ehp.10290](https://doi.org/10.1289/ehp.10290).
- 245 J. Li, *et al.*, Toxicity of inorganic nanomaterials in biomedical imaging, *Biotechnol. Adv.*, 2014, 32(4), 727–743, DOI: [10.1016/j.biotechadv.2013.12.009](https://doi.org/10.1016/j.biotechadv.2013.12.009).
- 246 K. Donaldson and C. A. Poland, Nanotoxicity: challenging the myth of nano-specific toxicity, *Curr. Opin. Biotechnol.*, 2013, 24(4), 724–734, DOI: [10.1016/j.copbio.2013.05.003](https://doi.org/10.1016/j.copbio.2013.05.003).
- 247 A. J. Beer, *et al.*, Non-invasive assessment of inter- and inpatient variability of integrin expression in metastasized prostate cancer by PET, *Oncotarget*, 2016, 7(19), 28151–28159, DOI: [10.18632/oncotarget.8611](https://doi.org/10.18632/oncotarget.8611).
- 248 R. Bendardaf, *et al.*, Comparison of CD44 expression in primary tumours and metastases of colorectal cancer, *Oncol. Rep.*, 2006, 16(4), 741–746.
- 249 F. Fontana, *et al.*, Transferrin receptor in primary and metastatic breast cancer: evaluation of expression and experimental modulation to improve molecular targeting, *PLoS One*, 2023, 18(12), e0293700, DOI: [10.1371/journal.pone.0293700](https://doi.org/10.1371/journal.pone.0293700).



- 250 C. Chen, S. Zhao, A. Karnad and J. W. Freeman, The biology and role of CD44 in cancer progression: therapeutic implications, *J. Hematol. Oncol.*, 2018, **11**(1), 64, DOI: [10.1186/s13045-018-0605-5](https://doi.org/10.1186/s13045-018-0605-5).
- 251 H. He, L. Liu, E. E. Morin, M. Liu and A. Schwendeman, Survey of Clinical Translation of Cancer Nanomedicines-Lessons Learned from Successes and Failures, *Acc. Chem. Res.*, 2019, **52**(9), 2445–2461, DOI: [10.1021/acs.accounts.9b00228](https://doi.org/10.1021/acs.accounts.9b00228).
- 252 H. Tao, T. Wu, M. Aldeghi, T. C. Wu, A. Aspuru-Guzik and E. Kumacheva, Nanoparticle synthesis assisted by machine learning, *Nat. Rev. Mater.*, 2021, **6**(8), 701–716, DOI: [10.1038/s41578-021-00337-5](https://doi.org/10.1038/s41578-021-00337-5).

