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Basic Study on Gene Biology of Bladder Cancer Metastasis

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Abstract

Bladder cancer is a complex disease with distinct treatment approaches based on its progression. For non-muscle invasive bladder cancer, the primary treatment method involves complete tumor resection, followed by immunotherapy, intravesical chemotherapy, and regular monitoring. In cases of muscle-invasive bladder cancer, a multimodal approach-including radical cystectomy and neoadjuvant chemotherapy offers the best chance of cure; though some tumors still progress to metastatic disease, which is associated with high mortality rate. Metastasis remains the primary cause of bladder cancer mortality. Since research on tumor metastasis began in 1889, discoveries like the seed and soil hypothesis and the role of host factors have shaped the treatment strategies and contributed to our understanding of metastatic behavior. Advancing our knowledge of tumor biology, particulary in relation to metastasis, remains essential, and summarizing current findings in this area will support further progress in bladder cancer metastasis research.

Key words bladder cancer, tumor metastasis, basic research, biology

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Introduction

Metastasis refers to the process where transformed tumor cells, originating from the primary site, migrate through blood vessels or lymphatic vessels to new tissue environments and initiate clonal growth. This is a dynamic, multifaceted, and complex biological process that involves interactions between tumor cells and the host organ microenvironments. Over more than a century of metastasis research (**Figure 1**), several fundamental principles have emerged: 1. Tumors exhibit biological heterogeneity; 2. The metastatic process is highly selective, with only a small subset of cells from diverse primary tumors possess the ability to survive and proliferate; 3. The success of metastatic growth depends on the complex interactions between metastatic cells and the stromal cells within the host microenvironment they invade. The metastatic steps can be described as follows: 1) Tumor cell migration, gradually detaching from the primary site; 2) Invasion into neighboring tissues and penetration through the basement membrane; 3) Entry into the blood or lymphatic vessels; 4) Tumor cells evading apoptosis, allowing survival within the circulatory system; 5) Extravasation from blood or lymphatic vessels into distant organs or tissues; 6) Formation of micro-metastatic niche; 7) Adaptation to the surrounding microenvironment, leading to the establishment of metastatic lesions. Metastasis is the most fundamental malignant feature of malignant tumors, and the control of metastasis is the key to treatment [1, 2].

Urothelium is the epithelial tissue that lines the entire urinary tract, including the renal pelvis, ureters, bladder, and proximal urethra. Bladder cancer is the most common tumor in the urothelium. The bladder serves as a storage organ for urine and acts as a barrier between urine and blood, and it is frequently exposed to various potential carcinogens [3]. Compared to other types of tumors, bladder cancer exhibits distinct characteristics of progression through various pathways that include Non-Muscle Invasive Bladder Cancer (NMIBC), accounting for 70-80% of bladder cancers, is typically of low grade, superficial, and papillary. Approximately 70% of these tumors recur, but only about 15% progress to muscle invasion. These tumors often harbor mutations in the HRAS gene (30-40%) and the Fibroblast Growth Factor Receptor (FGFR3) gene (~70%), indicating that the activation of RTK-Ras plays an early and crucial role in this pathway of tumorigenesis. Muscle-Invasive Bladder Cancer (MIBC), which accounts for 20-30% of bladder cancers, is characterized by high-grade muscle-invasive tumors. These tumors may either originate from flat carcinoma in situ (CIS) or present high-grade and invasive features from the onset of tumorigenesis. More than half of MIBC cases contain structural and functional defects in tumor suppressor factors such as p53 and/or retinoblastoma protein (RB), with over 50% progressing to local and distant metastasis [4]. In bladder cancer, MIBC (muscle-invasive subtype) is the major cause of bladder cancer-related deaths. The five-year survival rate for MIBC drops from less than 50% in the absence of metastasis to 5% in cases with distant metastasis, indicating that metastatic factors in MIBC are the primary contributors to bladder cancer deaths. Studying the biological processes of bladder cancer cell metastasis is of great value for identifying potential therapeutic targets and improving the prognosis of bladder cancer patients [5].

One crucial lesson from humanity's long battle against disease is that only by deepening our understanding of disease mechanisms can we develop more effective treatment strategies. Currently, effective cancer treatments are based on a deeper understanding of tumor biology, with the development and use of targeted therapies serving as a prime example of this approach. Among these, a notable example is the development of Imatinib (Gleevec). Through the exploration of disease mechanisms, researchers ultimately discovered that the BCR-ABL fusion gene is the cause of chronic myeloid leukemia (CML). Through ongoing design and refinement of the drug molecules, researchers successfully developed Imatinib mesylate, transforming CML into a manageable chronic disease. Similarly, by gaining a thorough understanding of the mechanisms that hinders the immune system's ability to eliminate tumor cells, scientists developed immune checkpoint inhibitors, greatly enhancing the immune system's ability to target and destroy tumors. For example, antibodies and small molecule inhibitors based on PD-1/PD-L1, as well as Chimeric Antigen Receptor T-cell Immunotherapy (CAR-T), were developed. In conclusion, understanding the mechanisms behind tumor metastasis is essential for developing more effective strategies to combat it. This review will explore bladder cancer metastasis from a researcher's perspective, examining various key aspects of this complex process.

Research models and tools for bladder cancer metastasis

Syngeneic tumor model

There are mainly two types of models: syngeneic transplantation models and transgenic mouse models. The former refers to the inoculation of tumor cells or tissues from the same species, such as mice, into the subcutaneous tissue, vascular system, or primary tumor site, to observe the tumor growth, progression, metastasis, and other processes. In this model, both the graft and the host belong to the same species, and the host retains normal immune function. Another method is carcinogen-induced models In bladder cancer research, the most common spontaneous tumor model is the BBN-induced mouse bladder cancer model. However, the heterogeneity of the cancer phenotype makes it difficult to use in molecular research or preclinical studies, and it rarely forms metastases [6, 7].

Transgenic mouse models primarily refer to models that develop tumors spontaneously through gene modification, leading to progression and metastasis. For instance, the MMTV-polyoma virus middle T antigen (PyVmt) transgenic mouse is a model often used in breast cancer research. Through gene modification and exposure to carcinogens, tumors naturally develop in specific organs or sites within the mouse, subsequently metastasizing to distant organs. Transgenic mice maintain relatively intact immune system functions, making the research results more reflective of real-world scenarios. Compared to other cancers, the availability of bladder cancer models is somewhat limited, with the representative nature of genetically engineered mouse (GEM) models being notably deficient [8]. Transgenic spontaneous bladder cancer models, for example, involve the expression of oncogenes in the urinary epithelial cells of mice through genetic modification. Germline models, such as the construction of P53 knockout mice or mice with conditional knockout of tumor suppressor genes, are also included. However, these models have the following disadvantages: 1. Limited selection of genes that are specifically expressed in the bladder; 2. The phenotypes are relatively mild; 3. There are very few models that develop invasive bladder cancer; 4. Metastasis is rarely observed [9-11].

Xenograft models

Primarily, human tumor cells or fragments are implanted into immunodeficient experimental animals. It is clear that one significant disadvantage of this model is the inability to observe the role of the immune system in tumor progression and metastasis [12]. Nevertheless, the xenograft model has been proven to be valuable for understanding cancer biology and metastasis due to its relatively short duration and low cost, making significant contributions to the study of tumor metastasis. Furthermore,

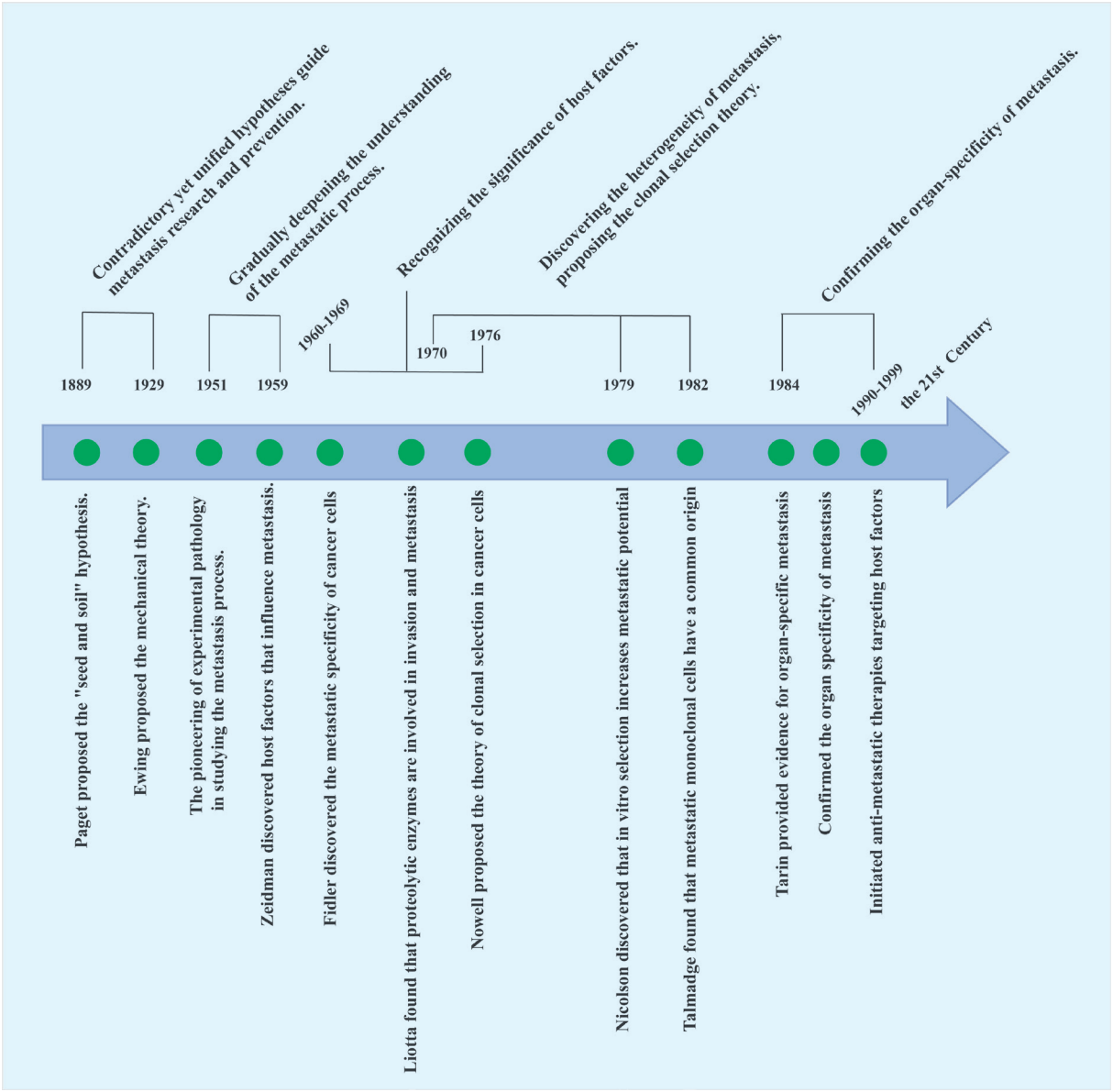

Figure 1. Timeline of tumor metastasis research. This timeline represents a historical overview of the significant milestones and discoveries in the field of tumor metastasis research, highlighting key breakthroughs that have shaped our understanding of the metastatic process.

the application of orthotopic implantation techniques more closely resembles the complex tumor microenvironment and growth, providing a more comprehensive simulation of the entire metastasis process [8]. It is worth noting that the orthotopic bladder cancer model has not been widely used due to the thin bladder wall and the large volume of the free portion. Gabri van der Pluijm and colleagues utilized poly-L-lysine to damage the bladder wall while retaining perfusion of luciferase-labeled tumor cells, allowing for a comprehensive and efficient simulation of the entire process of bladder cancer progression and distant metastasis. This approach offers higher cost-effectiveness and practicality, making it an ideal preclinical model [13]. Furthermore, the PDX (Patient-Derived Xenograft) mouse model, which involves implanting a patient's tumor into recipient mice, also falls under the category of heterologous models. The tumor in PDX mice is directly derived from the patient, providing a clear advantage in selecting new drugs after the tumor develops drug resistance. An increasing number of PDX models are rapidly replacing long-established traditional cell lines, becoming the preferred model for conducting basic and translational preclinical research [14].

The study of classical tumor metastasis suppressor genes and promoter genes in bladder cancer

In the study of tumors, it is established that proto-oncogenes play a dominant role in tumorigenesis and progression. However, incidence data did not support this hypothesis. Subsequently, researchers discovered and proposed a large subset of genes that play a crucial role in tumorigenesis—tumor suppressor genes, such as RB, P53, APC, PTEN, and TSC1. The loss of function of these genes is critical for tumor development. Identification of oncogenes and tumor suppressor genes provides a simple yet powerful explanation for the occurrence and progression of tumors [15]. Tumor metastasis is a complex, inefficient yet deadly process. Completion of metastasis requires coordination of the activation of metastasis-promoting genes and the inactivation of

metastasis-inhibiting genes within the tumor, as well as the tumor microenvironment that permits escape of cancer cells from the primary site and growth in secondary organs. Therefore, in 1988, Patricia and her colleagues proposed a hypothesis that, similar to tumor suppressor genes, there exist metastasis suppressor genes (Metastasis Suppressor Genes), the loss of which imparts cells the ability to metastasize.

Tumor metastasis suppressor genes

The definition of metastasis suppressor genes highlights their important role in inhibiting the formation of metastases. The earliest successful technique used for the screening of metastasis suppressor genes (MSG) was the MMCT method. Specifically, this involves inhibiting the mitosis of growing cells, allowing chromosomes to drift naturally, and then encapsulating the drifting chromosomes into new micronuclei. Subsequently, employing methods such as altering their chemical properties, differential centrifugation, and filtration, microcells containing a single chromosome were selected. These microcells are then fused with tumor cells to form new hybrid cells, in order to screen for MSG. Although the efficiency is low and the steps are complex, through the persistent efforts of previous researchers, genes such as BRMS1 have been identified [16]. With the advancement of high-throughput technologies such as chips and sequencing, a series of MSGs have been identified and validated (**Table 1**). Except for a few genes like RhoGDI2 and CDH1, most genes in the table lack direct experimental evidence for their role in inhibiting bladder cancer metastasis. By elucidating the roles and mechanisms of these genes in bladder cancer metastasis, it is hoped that potential therapeutic targets with application value can be discovered.

Oncogene promoting tumor metastasis

Receptor tyrosine kinase (RTK). Receptor Tyrosine Kinases (RTKs) are a family of cell surface receptors, with 58 known RTKs in humans, which can be divided into 20 subfamilies [53]. RTKs serve as receptors for growth factors, hormones, cytokines, neurotrophic factors, and other extracellular signaling molecules. RTKs mediate key signaling pathways involved in cell proliferation, differentiation, survival, and cell migration, where the activation mutation or overexpression of these molecules plays an important role in tumor development and progression. Considering EGFR as an example, when the ligand binds to it, the bound EGFR receptor undergoes a conformational change in its extracellular domain, leading to dimerization with other receptors of the same or similar family members. This dimerization results in the mutual phosphorylation of specific sites on EGFR, thereby activating the receptor. These phosphorylated sites serve as docking points for various scaffolding proteins or kinases originally located in the cytoplasm, facilitating their translocation to the membrane and thereby activating a series of downstream pathways that regulate cell behavior. These scaffolding proteins include PKC, PI3K, RAS, SRC, ABL, PAK and STAT5 [53, 54].

EGFR. More than 50% of tumors exhibit high expression of EGFR, and its expression level is directly related to tumor grade, stage, and survival rates. In muscle-invasive bladder cancer (MIBC), overexpression of EGFR is associated with lower tumor-specific survival. The relationship between EGFR and the invasion and metastasis of bladder cancer is also reflected in preclinical models: compared to 253J cells, 253JBV cells (which are highly metastatic cells screened after continuous passage in mice) exhibit overexpression of EGFR [55, 56]. Similarly, other experiments have confirmed that EGFR promotes metastasis by enhancing the proliferation, angiogenesis, and invasion of bladder cancer cells. EGFR family consists of four members: in addition to the previously mentioned EGFR, there are human EGFR 2, human EGFR 3, and human EGFR 4. EGFR2 cannot form homodimers; instead, it functions by forming heterodimers with other EGFR members, which then transmit signals downstream to regulate cellular behavior [57]. Expression of EGFR2 positively correlated with the metastatic capacity of MIBC [58]. Although there are studies suggesting that EGFR3 and EGFR4 may play significant roles in the progression of bladder cancer, there is no direct evidence demonstrating that EGFR3/4 directly promotes bladder cancer metastasis. EGFR can promote bladder cancer metastasis by regulating the EMT (epithelial-mesenchymal transition) process. Preclinical studies on other cancer models have identified that EGFR can promote EMT through STAT3 signaling, suggesting that high levels of EGFR expression may likely contribute to EMT in basal-like bladder cancer [59].

Matrix metalloproteinases (MMPs). Matrix metalloproteinases (MMPs) are a family of proteolytic enzymes that function extracellularly by altering the cellular microenvironment. Their substrates include basement membranes, extracellular matrix (ECM) molecules, and various extracellular cytokines, indicating that MMPs play a central role in normal physiological processes, benign diseases, and malignant tumors [16]. MMPs are crucial for tumor-induced angiogenesis, tumor invasion, and the establishment of metastatic foci at secondary sites [60]. Tumor cells must detach from the primary site, degrade the extracellular matrix (ECM), and invade the stroma as a prerequisite to enter the bloodstream and colonize at distant sites. Moreover, at the secondary sites, tumor cells must induce and establish a blood supply to create conditions favorable for proliferation [61, 62]. Although invasion and angiogenesis are very different processes, both invasion and angiogenesis require proteases such as MMP-9 to alter the extracellular matrix and basement membranes [63]. Studies have shown that the mRNA expression levels of MMP2 and MMP9 are higher in muscle-invasive bladder cancer (MIBC) than in non-muscle-invasive bladder cancer (NMIBC), and their high expression is negatively correlated with bladder cancer survival rates, indicating prognostic value in MIBC [64, 65]. Moreover, their roles in the proteolytic processing of various growth factors, growth factor receptors, and cytokines have been recognized. In a different mouse model, after intracardiac injection of bladder cancer cells, continuous retrieval of bone metastatic foci revealed increasing levels of MT1-MMP, MT2-MMP, MMP-9 and TIMP-2, which were associated with metastatic potential. The use of MMP-2 and MMP-9 gene knockout mice further reinforced the importance of these genes in metastasis [67, 68]. Notably, in MMP-9 gene knockout mice, there was a significant reduction in spontaneous metastasis compared to wild-type mice, using different types of tumors [66].

RHO family genes. Small GTP-binding protein Rho and its most typical downstream effector, Rho-associated serine/threonine protein kinase (ROCK), are involved in actin cytoskeletal organization and are associated with the pathogenesis and progression of several human cancers [69, 70]. Members of the Rho subfamily of small GTPases are involved in regulating various cellular processes, including microfilament organization, cell-to-cell contact, and malignant transformation of cells [70]. Indeed, these events are also interrelated. Specifically, the Rho subfamily regulates the formation of intracellular stress fibers and focal adhesions. The Rac subfamily regulates the formation of lamellipodia and membrane ruffles, while the Cdc42 subfamily regulates the formation of filopodia [71, 72]. Lamellipodia and filopodia appear on the leading edge of motile cells, while retraction occurs on the trailing edge [73]. These changes are the basis of cell migration. Cancer cell migration is central to the metastatic process. The molecular alterations described above, resulting in corresponding changes in cell migration ability, are

Table 1. Tumor metastasis suppressor genes.

Gene	As evidence for metastasis suppressor genes	References
CDH1	Cell-cell/cell-matrix adhesion.	[17]
CDH11	Knockdown inhibits EMT and tumor cell stemness.	[18, 19]
CRSP3	Overexpression exogenously can inhibit metastasis.	[20]
KISS1/KISS1R	Exogenous overexpression of KISS1 allows cells to disseminate, but they do not proliferate in the secondary organs.	[20, 21]
KLF17	Overexpression inhibits metastasis; knockdown promotes EMT transformation.	[22]
MAK7	Regulating the MAPK pathway affects metastasis.	[23]
AKAP12	Functioning through JN and/or Raf/MEK/ERK.	[24]
BRMS1	In experimental metastasis analysis, reduced tumor metastasis rate and size.	[25]
Caspase 8	Induced apoptosis/anoikis.	[26]
CD44	Cell-cell/cell-matrix adhesion.	[27, 28]
Claudin-4	Inhibiting anchorage-independent growth processes.	[29]
CTGF	Lung metastatic lesions size reduction by 15%-25%.	[30, 31]
DCC	Cell-cell/cell-matrix adhesion; Apoptosis.	[32]
DLC1	After re-expression, significant inhibition of tumor metastasis.	[33, 34]
DRG1	Overexpression of Drg-1 in metastatic colon cancer cells reduces in vitro invasion through Matrigel and inhibits liver metastasis in nude mice.	[35, 36]
GAS1	GAS1 inhibits the glycolytic process, thereby suppressing tumor metastasis.	[18]
Gelsolin	Affecting the cytoskeleton affects metastasis.	[37, 38]
KAI-1	Binding to DARC on the surface of vascular endothelial cells induces tumor cell growth arrest.	[39]
LSD1	The influence on the cytoskeleton affects metastasis.	[40]
MAP2K4	Constitutively active MAP2K4 increases tumor size and the number of circulating tumor cells in the blood and bone marrow.	[41]
MKK-4	Overexpression subsequently inhibits the metastasis process.	[42]
MKK-7	Overexpression suppresses the metastatic process.	[43]
Nm23	Inhibition of metastatic clone growth.	[44]
OGR-1	Inhibition of the metastasis process.	[45]
PEBP1	Referred to as Raf kinase inhibitor protein (RKIP).	[46]
RhoGDI2	Reduce tumor metastasis rate and size; it is an important target of the anti-angiogenic compound apatinib.	[47]
RKIP	Regulate the expression of angiogenic genes; regulate the checkpoint of the spindle in the cell cycle.	[48]
RRM1	Inhibit tumor metastasis by positively regulating PTEN.	[49]
SMAD7	Overexpression of Smad7 delays the establishment and growth of a mouse model of melanoma bone metastasis.	[50]
SSeCKS	Reducing the rate and size of tumor metastasis.	[51]
TXNIP	Influencing metastasis by regulating cellular redox homeostasis.	[20, 52]

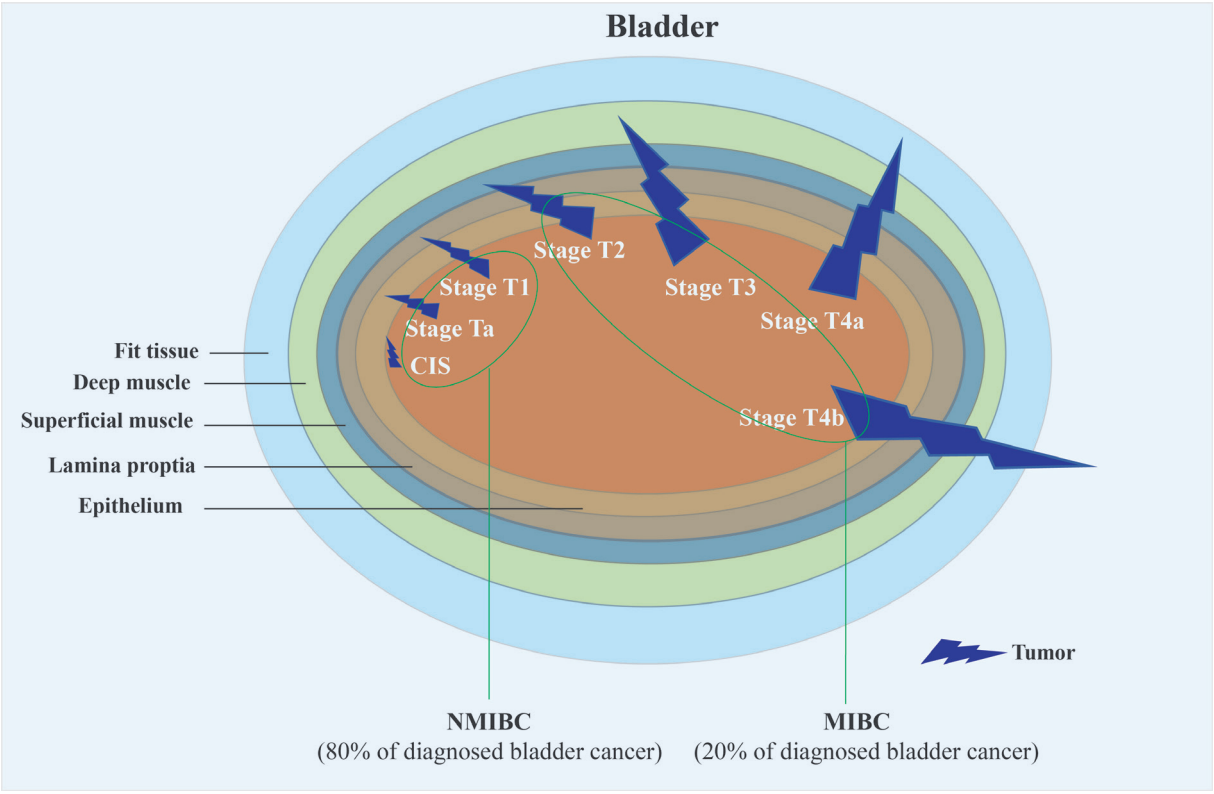

Figure 2. The TNM staging system for bladder cancer is a standardized method used by healthcare professionals to classify and categorize the extent of the cancer based on three key components: Tumor (T), Nodes (N), and Metastasis (M). This system helps in determining the stage of the cancer, which in turn influences treatment options and prognostic assessments.

crucial for the in vivo metastasis of tumor cells. Rho proteins act as molecular switches inside cells, transmitting extracellular stimuli signals to the actin cytoskeleton and nucleus, regulating cell migration and malignant transformation. The rho/rock signaling pathway is involved in the regulation of the cytoskeleton, thereby affecting various behaviors such as cell migration and invasion, promoting tumor metastasis. By regulating the RhoA/ROCK signaling pathway, MALAT1 promotes the metastasis of osteosarcoma [74]. AFAP1-AS1 may promote the metastasis of nasopharyngeal carcinoma cells by regulating tumor cell adhesion and migration through the RhoA/Rac2 signaling pathway [75]. In the previous section on tumor suppressor genes, it is mentioned that one of the key tumor suppressor genes identified in bladder cancer is RhoGDI2, whose full name is Rho GDP dissociation inhibitor 2. This gene exerts its function by binding to Rho family members and inhibiting their activity. Researchers have found that reactivation of RhoGDI2 can effectively prevent the occurrence of bladder cancer invasion and metastasis events [76]. These results indicate that some members of the Rho family are key factors in promoting bladder cancer metastasis. Additionally, MMP-2, as a downstream molecule of RhoGDI2, is positively regulated by it and plays a role in the invasion of bladder cancer into surrounding tissues and the metastatic process [77].

Epithelial-mesenchymal transition (EMT) and bladder cancer metastasis

EMT is the phenomenon of epithelial-mesenchymal transformation, which was first proposed by Greenberg in 1982. Based on the specific biological context in which EMT occurs, it can be divided into three subtypes. Type 1 and Type 2 EMTs refer to EMTs associated with embryo implantation, development, and organ formation, and those related to tissue regeneration, injury repair, and organ fibrosis, respectively. Type 3 EMT refers to the process in which epithelial-like cells transform into mesenchymal-like cells during tumor development [78]. EMT is related to the drug resistance and metastasis of bladder cancer cells and is a hot area of research in bladder cancer [79]. The reciprocal conversion mechanism of EMT and MET enables tumor cells to go through various stages during metastasis, such as migrating out of the primary site and into the secondary site, surviving in the blood circulation, proliferating in the secondary site, and inducing angiogenesis, as the biological foundation [80]. Specifically, inflammatory cytokines [81] and the extracellular matrix trigger EMT in subpopulations of cells within the primary tumor, allowing them to collectively invade and enter circulation [82]. Subsequently, tumor cells interact with platelets, which provide TGF, thereby enhancing EMT and allowing migration to distant organs and invasion [83]. At this point, EMT is reversed [84], as the epithelial phenotype appears to enable faster tumor cell proliferation compared to maintaining a mesenchymal state.

The whole-genome mRNA expression profiles of primary bladder cancer and circulating tumor cells (CTCs) in bladder cancer show that tumor cells in the bloodstream have higher expression levels of EMT markers, which is consistent with observations in other cancer models [83-85]. Notably, CTCs exhibit increased expression of SNAI1 (Snail), and conditional knockout of SNAI1 blocks the production and metastasis of CTCs. When exosomes

from MIBC cell lines stimulate primary urothelial carcinoma cells (**Figure 2**), the urothelial cells undergo EMT, accompanied by enhanced cell migration abilities [86]. At the molecular level, EMT is characterized by the loss of E-cadherin and increased expression of several transcriptional repressors of E-cadherin. A significant feature of bladder cancer is its development along two seemingly distinct pathways at the phenotypic and molecular levels. Researchers have conducted clustering analysis on the expression of EMT markers (E-cadherin, Zeb-1, and Zeb-2) and the "dual-track" developmental pathways exhibited by bladder cancer. The results confirmed that the expression of Zeb-1 and Zeb-2 is highly enriched in MIBC, supporting the concept that EMT may serve as a basis for the invasion and metastasis of bladder cancer. These findings indicate that EMT plays a crucial regulatory role in the invasion/metastasis process of various tumors, including bladder cancer, and it is also a major characteristic of the entire cancer biology.

Discussion

When tumor cells leave the primary tumor site, they typically face three outcomes in the target organ: 1) death, 2) proliferation, and 3) dormancy. Therefore, one of the potential factors contributing to the organ selectivity of tumor metastasis [15] is the result of tumor dormancy in certain organs. As observed through long-term studies, breast or prostate cancer often metastasizes to bone, while gastric or colon cancer is more prone to liver metastasis. The underlying reason may be that the dissemination of tumor cells is random, and whether macroscopic metastatic foci are formed depends on whether the "seed" (tumor cells) and "soil" (organ environment) are compatible. If the environment is suitable for growth, tumor cells can wake up from dormancy and form metastatic foci. Dormant tumor cells exhibit the following characteristics: 1) Reduced cellular activity; 2) Greater stem-like properties; 3) Greater resistance to treatments targeting rapidly proliferating tumors; 4) Clinical difficulty in identifying the existence of dormant cells; 5) Benign-like state; 6) Difficulty in studying in vitro environments [87-89]. Therefore, conducting relevant research remains fraught with challenges. Despite this, researchers have made some significant findings.

In the earlier chapters of this review, we introduced MSGs—metastasis suppressor genes. These genes are unified by their ability to inhibit metastasis. As researchers explore the mechanisms by which MSGs prevent metastasis, they have found that MSGs play a significant role in promoting the dormancy of disseminated tumor cells, which is one of their key functions [90]. RhoGDI2 has been defined as a gene that inhibits metastasis without affecting the proliferation of primary tumors. Its mechanism may involve inducing or maintaining tumor cells in a dormant state [91]. Evidence suggests that the metastasis suppressor factor RhoGDI2 inhibits the endothelin axis and interacts with macrophages in the microenvironment of micrometastasis, thereby suppressing metastatic growth.

Another MSG we mentioned earlier, KISS1, is a ligand for the G protein-coupled receptor (KISS1R) and exerts a strong inhibitory effect on metastasis across various tumor types, including breast cancer, melanoma, prostate cancer, ovarian cancer, and pancreatic cancer. Studies in these tumors suggest that the mechanism by which kiss1/kissR inhibits tumors may also involve mediating tumor cell dormancy [88]. Furthermore, the dormancy of tumors in target organs, while making metastasis appear inefficient, is not uncommon for cases where, years after the removal of the primary tumor, cells re-enter the cell cycle and proliferate under suitable conditions, leading to recurrence [88]. This indicates that research into this mechanism has potential clinical significance. In summary, the study of tumor dormancy poses a challenging issue. A deeper understanding of the mechanisms underlying this phenomenon will guide the development of new therapeutic strategies targeting tumor metastasis.

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Ethical policy

All procedures performed in this study were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards. Informed consent was obtained from all individual participants included in the study. Approval from institutional ethical committee was taken.

Availability of data and materials

All data generated or analysed during this study are included in this publication.

Author contributions

CJ searched academic literature, wrote the draft manuscript, made the figures and submitted the final manuscript.

Competing interests

Authors report no conflict of interest.

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