

Bacteria in the tumor ecosystem

From mechanisms to engineering

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Abstract: Many adjuvant factors surrounding the tumor aid nutrient capture and immune escape of the tumor cell and promote tumor growth, local spread, and distant metastasis. The tumor microenvironment (TME) consists of immune cells, cancer-associated fibroblasts (CAFs), endothelial cells, and the extracellular matrix (ECM). Recent studies have shown that bacteria can be present in the solid tumor and that they are an important part of the TME. In this review, we explore the contribution of bacteria to developing the TME and analyze systematically their role in cancer initiation and progression. In addition, we discuss antitumor immunotherapies that target bacteria inside the tumor. This review considers the application of the tumor-specific delivery systems developed via bacterial bioengineering in antitumor therapy. We also discuss future trends and challenges for the use of bacteria in antitumor therapy. The unique contribution of this review lies in our systematic analysis of how bacteria reshape the tumor ecosystem through the bacteria–matrix–immune tripartite interaction, in addition to compiling evidence of their presence.

Abbreviations: CAFs = cancer-associated fibroblasts, ECM = extracellular matrix, EMT = epithelial–mesenchymal transition, ICD = immunogenic cell death, MDSCs = myeloid-derived suppressor cells, NK = natural killer, OS = overall survival, PFS = progression-free survival, TAMs = tumor-associated macrophages, TME = tumor microenvironment, TLRs = Toll-like receptors, VEGFs = vascular endothelial growth factors

Keywords: bacteria, engineered bacteria, extracellular matrix, immunotherapy, tumor microenvironment

1. Introduction

Tumors are highly complex diseases not only generated from genetic mutations and phenotypic modifications occurring within cancer cells, but are also greatly conditioned by the tumor microenvironment (TME) in which they develop. However, the tumor is not just a pack of cancer cells, but a constantly changing ecosystem encompassing heterogeneous components, comprising cancer cells, immune cells, blood vessels, cancer-associated fibroblasts (CAFs), extracellular matrix (ECM), extracellular molecules, and microorganisms. Inside this system, these features interact with each other via complex signaling pathways that together generate a microenvironment that stimulates tumor growth, impedes cancer immunity, and allows distant

tumor metastases. Traditional oncology research focused on cancer cells and studied the host immune system's surveillance and defense functions. But accumulating evidence now suggests that all types of cells involved in cancer also have critical functions in the evolution of tumors.

For example, CAFs facilitate tumor invasion by secretion of growth factors and by remodeling the ECM^[1,2]; tumor-associated macrophages (TAMs) often have a pronounced immunosuppressive phenotype that facilitates tumor immune escape^[3,4]; and aberrant angiogenesis favors chronic tumor hypoxia and tumor-cell lactate accumulation.^[5–7] Together, these factors lead to a severely immunosuppressive context that favors tumor progression.

The presence of low-biomass microbial communities within tumors has historically been a subject of intense debate. Early scholars suggested that detected bacterial signals might stem from exogenous contamination or post-biopsy artifacts rather than intrinsic tumor components. However, with the advent of high-precision sequencing technologies and stringent contamination control protocols, such as employing blank controls during DNA extraction and implementing computational filtering to remove environmental and reagent-derived sequences, the presence of tumor-associated microbial communities has been unequivocally validated. Of late, one remarkable finding has been that bacteria can be important players in the TME. Already in the early part of the last century, several bacterial infections were identified as being causative of cancer. For

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instance, infection with *Helicobacter pylori* (*H. pylori*) is a major causative factor in gastric cancer. However, most of these studies were limited to cancers associated with the mucosa, and for decades, most workers presumed that solid tumors (with their paucity of blood vessels and absence of open passages) were unlikely to contain stable bacterial communities. Recent technical advances in high-throughput sequencing and single-cell spatial omics studies have slowly challenged this notion. Growing evidence now indicates the presence of diverse bacteria in breast, bone, pancreatic, colorectal, and lung cancers and also in melanoma. Such bacteria not only colonize tumors but also invade deep within tumor tissue, and in some cases, live inside the cytoplasm of cancer cells.^[8,9] Functions of bacteria in the TME are diverse and intricate. On the one hand, some bacteria support the growth and spread of cancer cells via their metabolites or immune regulation. For example, the bacterium *Fusobacterium nucleatum* (*F. nucleatum*) binds to host cell receptors through its adhesins to activate several proinflammatory signal pathways that increase tumor cell invasiveness^[10] and, at the same time, influences T-cell and dendritic cell function by suppressing them, thus reducing host immune defense.^[11,12] Conversely, there are bacteria exerting a beneficial effect in anticancer therapy. Some probiotics, including *Lactobacillus* and *Bifidobacterium*, have been demonstrated in many studies to modulate immune responses and boost antitumor immunity.^[13,14] In addition, some anaerobic bacteria exhibit natural tumor tropism, enabling them to specifically colonize hypoxic necrotic tumors and to thus serve as a basis for engineering bacteria into therapeutic carriers with tumor homing capability.^[15,16] There have been rapid advances in this field such that “bacteria within the tumor environment” is rapidly emerging as a new research focus area in cancer research. In addition to describing their presence in the local environment, an increasing number of studies are starting to explore bacterial modes of action to tumorigenesis and progression, as well as options for targeting intratumoral bacteria therapeutically. For example, some reports propose using antibiotics or bacteriophages to kill tumor-associated carcinogenic bacteria to improve the effect of immunotherapy or chemotherapy; others use synthetic biology to synthesize bacteria to carry drugs or immune-modulating substances, to actively deliver to tumors, to precisely and efficiently treat cancer. However, studies into the interactions of bacteria with tumors still have major challenges ahead. First, bacterial loads in tumors are low and much prone to experimental contamination, thus methodological refinements to accurately quantify and analyze bacterial communities associated with a tumor are required. Second, mechanisms involved in host immune system–bacterial interactions are complex and variable, and the same bacterial species may exert different effects on cancer types or on the same patient. Third, facilitating precise bacterial-targeted therapy approaches in clinics and preserving the resident bacteria are challenging issues for the future.

Against this background, this review not only compiled evidence of bacterial presence but also systematically

elucidated how they reshape the tumor ecosystem through the bacteria–matrix–immune tripartite interaction. First, the review lists the evidence of bacterium presence in various tumors and their distributions; then discusses how bacteria may influence tumor growth through metabolism, via their immunomodulatory roles and their interactions with other microenvironmental constituents; then assesses current tumor immunity immunotherapy treatment strategies and antibiotics interventions on intratumoral bacteria; the new methods and clinical approaches of engineered bacteria for antitumor therapy in recent years have also been introduced. Finally, the review highlights the future directions and challenges of bacteria-based tumor therapy, to provide guidance and viewpoints to researchers on bacteria-based tumor therapy.

2. The relationship between bacteria and tumors

2.1. Advances in microbial detection technology

As our understanding of the role that bacteria play within the TME improves, faster and better technologies of detection play a crucial role in related research. Early studies mostly used the traditional culture methods. However, these methods usually could not give enough and useful information, owing to the low abundance of bacteria in the tumor tissue, its common “dormant” state, or intracellular status. Subsequently, molecular biology techniques such as polymerase chain reaction (PCR) and 16S ribosomal RNA (rRNA) sequencing were also used to explore the tumor-associated microbial community. These methods have higher sensitivity and specificity than traditional culture techniques and so allow investigation of low-abundance microbiota found within tumors and documentation of links between specific bacterial strains and a range of cancers. Nevertheless, such methods cannot provide information on the spatial distribution and make it difficult to investigate the location of bacteria within the TME and host cell or host cell–bacteria interactions. During the past few years, the advent of high-throughput sequencing and multiomics technologies has significantly boosted this research area.^[17,18] Metagenomics, transcriptomics, and single-cell sequencing not only reveal the communities of microbial diversity within tumor tissues but also elucidate the microbial metabolic functions and activity states. But emerging techniques such as spatial transcriptomics and multiplex *in situ* hybridization techniques, for example, enable mapping of bacteria distribution and the host cell response in the same histological context at the same time, providing new insights into how bacteria interact with the dynamics of the tumor immune microenvironment. Furthermore, advances in imaging technologies have provided novel insights into bacteria located in tumors. Techniques such as fluorescence *in situ* hybridization (FISH), immunohistochemistry, laser confocal microscopy, and electron microscopy have been used to assess bacterial presence in tumor tissues and the ultrastructure of bacterial cells.^[19,20] These

approaches not only bypass the spatial resolution limitation of molecular detection but also offer direct evidence of interactions between bacteria, immune cells, and ECM molecules. Overall, continuing improvements in detection technologies have allowed researchers to characterize the presence, distribution, and function of bacteria in association with different types of tumors at different levels and scales. These are expectations and recent trends in integrations, like the combination of molecular detection and spatial localization, and functional validation to dissect the bacterial landscape in the TME in a systematic way. These achievements not only elucidated tumorigenic mechanisms but also created a good technical foundation for the later bacteria-based diagnosis and treatment strategies.

With the developments of detection technologies, many studies have more recently confirmed the existence of bacteria in many different solid tumors and their roles in tumor-associated biological functions (Table 1). Alongside colorectal cancer, in multiple independent cohort studies, the abundance of *F. nucleatum* has been found to be remarkably elevated, and its enrichment in tumor tissue is strongly associated with an inferior clinical outcome.^[27–29] The invasion of *F. nucleatum* binds adhesins to host cell receptors. They stimulate host signaling pathways, for example, Wnt/ β -catenin, which stimulates tumor cell proliferation and invasion.^[30–32] In addition to *F. nucleatum*, some gut microbiota, such as *Bacteroides fragilis* (*B. fragilis*) and *Escherichia coli* (*E. coli*), produce toxins or metabolites that damage DNA or cause inflammation, and thus promote colon cancer.^[33,34] Another aspect is that bacterial presence in breast cancer tumor tissue is also supported by several lines of evidence. Metagenomic sequencing study also found considerable variation in bacterial community composition in the breast tumors versus those in the normal breast tissues, with an altered proportion

of Proteobacteria and Firmicutes.^[35–37] In breast cancer tissues, *F. nucleatum* can be detected with high efficiency, suggesting a role in promoting metastasis either by affecting the immune environment or by enhancing the motility of cancer cells.^[12,38] At the same time, some studies show that the ability of some lactic acid bacteria to be protective resides in blocking oncogenic signaling activities or altering host immune response, thus suggesting that the roles of different microbiota in breast cancer can be dual.^[13]

Microbiome studies of pancreatic cancer showed unique features. Pancreatic tissue used to be described as a sterile niche. Recent studies show that gut bacteria can populate the pancreas along the bile ducts or through the bloodstream.^[39] Studies have found that the composition of the microbiome in pancreatic tumors is markedly different from that of healthy people overall, with the tumor microbiota showing lower diversity.^[40] Some studies have demonstrated that the pancreatic tumor microbiota can influence the immune state of the TME and suppress antitumor immunity, stimulating tumor progression. For instance, particular gram-negative bacteria activate Toll-like receptors (TLRs) signaling by lipopolysaccharide (LPS) and result in the recruitment of immunosuppressive cells and thus can influence the response of patients to immunotherapy.^[41] Furthermore, lung cancer has its microbiome community features. On the other hand, recently, studies found that the microbial community structure of lung cancers derived from smoking is different from that from nonsmoking individuals.^[42–44] Several studies reported that the abundance of Actinobacteria, Firmicutes, and certain anaerobes is exaggerated in lung cancer tissue and, thereby, these communities may favor the carcinogenesis of lung cancer by affecting the local inflammatory response and oxidative stress.^[45,46]

Notably, the microbiomes of bronchoalveolar lavage fluid and of tumor tissues from lung cancer patients are

Table 1
Bacterial evidence in different tumor types.

Tumor type	Bacterial species	Mechanism
Gastric cancer	<i>H. pylori</i>	CagA and VacA induce chronic inflammation, reactive oxygen species/reactive nitrogen species (ROS/RNS) accumulation, and oncogenic signaling (SHP2 [Src homology 2 domain-containing protein tyrosine phosphatase 2]/ERK [extracellular signal-regulated kinase], NF- κ B [nuclear factor kappa-light-chain-enhancer of activated B cells], STAT3 [signal transducer and activator of transcription 3]), promoting genomic instability and malignant transformation. ^[21]
Colorectal cancer	<i>F. nucleatum</i> , <i>E. coli</i> , <i>B. fragilis</i>	FadA activates β -catenin/Wnt signaling; Fap2 (Fusobacterium autotransporter protein 2) interacts with TIGIT (T-cell immunoreceptor with Ig and ITIM domains)/Gal-GalNAc to suppress antitumor immunity and enhance invasion and chemoresistance. <i>B. fragilis</i> toxin (BFT) activates NF- κ B and Wnt pathways, inducing inflammation and epithelial proliferation. ^[22]
Breast cancer	<i>F. nucleatum</i> , <i>Methylobacterium</i> , <i>Proteobacteria</i>	Certain oral/gut bacteria can colonize breast tumors and modulate immune activity (CD8 ⁺ T cells/NK [natural killer] suppression), altering metastatic potential and tumor metabolism. ^[23]
Lung cancer	<i>Actinobacteria</i> , <i>Streptococcus</i> , <i>Veillonella</i>	Local microbiota alter epigenetic regulation and inflammatory signaling, influencing immunity and sensitivity to immune checkpoint inhibitors. ^[24]
Pancreatic cancer	<i>Gammaproteobacteria</i>	Intratumoral bacteria can metabolize or inactivate chemotherapeutics (e.g., gemcitabine) and promote immunosuppressive microenvironments. ^[25]
Bone cancer	<i>Fusobacterium</i> , <i>Staphylococcus</i> , <i>Pseudomonas</i>	Bacterial components may modulate TLR (Toll-like receptor)/NF- κ B signaling, chronic inflammation, osteoclast activation, and extracellular matrix (ECM) degradation, potentially facilitating invasion and lung metastasis. ^[26]

broadly consistent, and the disordered local microbiomes are likely to be related to cancer onset and development.^[47,48] Furthermore, growing evidence of the presence of bacteria in other types of tumors, such as melanoma and ovarian cancer, now exists. For example, immune-driving bacterial species have been identified in melanoma tumors and may impact the outcome of immunotherapy through affecting antigen processing or cytokine production, among other ways.^[49] Studies of ovarian cancer showed that the microbial content in the pelvic microenvironment is intimately linked to chronic inflammation and cancerous precursor lesions.^[50] New discoveries on bacterial evidence in bone tumors (e.g., osteosarcoma), recently reported, broaden the horizon of data related to the tumor-associated microbiome. Bone was long thought of as an otherwise low microbial-load tissue, yet evidence from multiomics studies, as well as pathological evidence, suggests that bacteria might play an underestimated role in bone tumors. Conclusion from culture and molecular amplification analyses of bone tumor samples from patients who underwent surgical resection found that certain bacterial species could survive and be involved in the tumor immune microenvironment.^[9,51] Interaction of the gut microbiota with osteosarcoma should be considered as well.^[52] Microbiome dysbiosis results in symbiotic bacterium depletion and is an intrinsic host factor contributing to the development of bone metastases.^[53]

Overall, the distribution of bacteria within various types of tumors is highly heterogeneous, and the role of

bacteria in these variations ranges widely: in some cases, bacteria are pathogenic agents that can promote tumor development and metastasis, and in other cases, bacteria protect against tumor growth or boost tumor immune-reaction. Moreover, this complexity makes it obvious that the tumor-associated microbiota does not function as unidirectional “drivers”, but is a dynamic control that has effects conditioned on multiple aspects (e.g., microbiota composition, host immune state and composition, properties of the tumor tissue). Future large-scale multiomics studies related to bacterial properties in different types of tumors will lead to a better understanding of bacterial mechanisms toward cancer and new insights for clinical diagnosis and personalized treatment.

2.2. Spatial distribution characteristics of tumor microbiota

Others show that bacteria are not randomly disseminated in tumor tissue but show distinct histological and spatial specificity. Studies of bacterial localization in different types of solid tumors consistently support an observation that bacteria localize to hypoxic or necrotic tumor areas where distinctive local tumor microenvironmental conditions and deficient immunity expose the tumor locally for an appropriate habitat for anaerobic or facultative anaerobic bacteria to invade into (Fig. 1). For example, high-density clusters of *F. nucleatum* can be found in breast and colorectal cancers, and

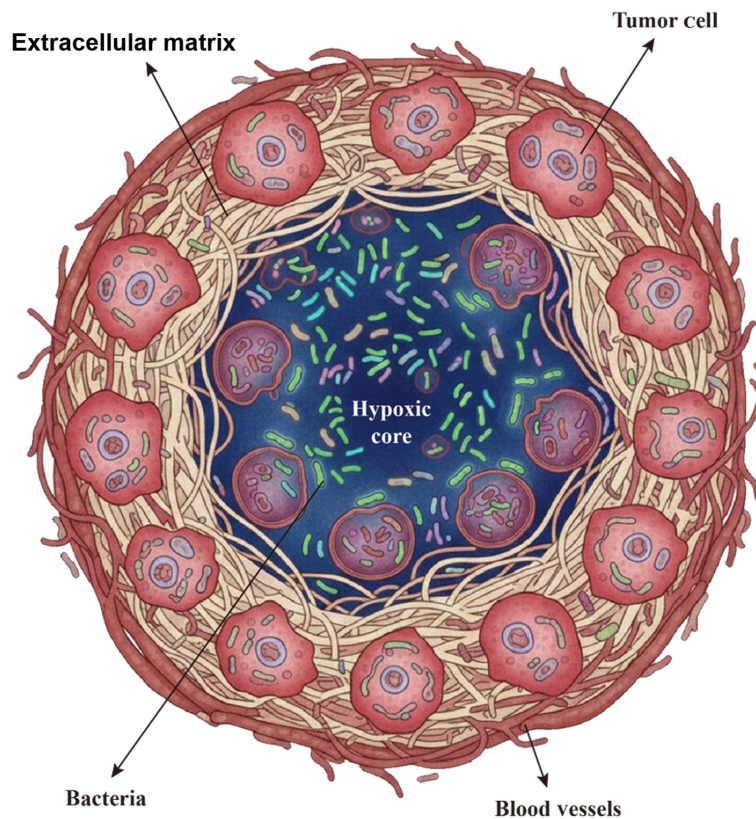


Figure 1. The spatial heterogeneous distribution of bacteria within tumors.

other bacteria can be found in pancreatic cancer and melanoma, mainly at the cores of tumors that have poor vascularization.^[8,38] But beyond spatial clustering, bacteria have intimate relationships with host cells. For example, some bacteria have been shown to invade and survive within cancer cells or tumor-associated immune cells and evade immune clearance, thereby providing an opportunity for bacteria to alter host cell signaling and activity through the secretion of secreted metabolites or virulence products. For instance, in pancreatic cancer tissues, bacteria can be seen within TAMs and CAFs, seemingly changing the functions of these cells in mediating immune responses and ECM production. Such intracellular colonization indicates that bacteria are not passive passengers of the TME but actively participate in the reshaping of the tumor environment by contacting and impacting major populations of cells within the tumor.^[41,54]

However, there are differences in patterns of bacterial presence among different types of tumors, related largely to the tissue of origin, vascularization, and immune barriers of tumors. For instance, the digestive system tumors would be more sensitive to the effect of intestinal microbiota translocation into tumors, and the tumors of some compartments, such as the skin or respiratory systems, may rely more on the microbiota of the local environment than other sites. The distribution patterns do have biological meanings and may also have clinical values. On the one hand, the spatial distribution and abundance of tumor-associated microbiota could act as diagnostic or prognostic markers to evaluate the risk of tumor growth. On the other hand, the characteristics of bacterial colonization can constitute an argument for bacteria-based targeting strategies, for example, by colonizing engineered bacteria in hypoxic areas within tumor tissue to deliver drugs therapeutically to specific locations. In summary, intratumoral bacterial abundance is not random and is jointly regulated by the tumor metabolic status, immune context, and organ-specific traits. A clear view of these distributions both explains how bacteria can contribute to the development and progression of tumors and provides crucial information that can aid in the development of novel diagnostics and therapeutic approaches.

2.3. The influence of bacteria on the TME

Cancer TME is the main scene of cancer occurrence and development, with hypoxia and acidosis, high concentration of lactic acid, and immunosuppressive conditions. The matrix involves numerous bacteria that have been only recently recognized as key components to participate in this environment; the bacteria are not passive elements. Rather, they profoundly affect tumor biological behavior at multiple levels through metabolic function, immune activities, and tumor stroma interactions.^[9]

2.4. The role of bacterial metabolites

Besides targeting the cancer cells directly, bacterial metabolites have an extremely complex and decisive role in cancer initiation and promotion. The bacterial metabolites can either induce tumor initiation or be antitumor in certain circumstances. Various bacterial communities of diverse contents affect the host cell to produce a variety of metabolites, impacting the stability of DNA, stimulating the activation of signaling pathways and immune response status, and so modulating the TME and guiding the disease course.^[55,56] Regarding the protumorigenic effect, bacterial metabolites might have protumorigenic effects, for example, by contributing to the tumorigenesis and invasion of tumors in a variety of different ways. For example, in hypoxic and metastable tumor surroundings, butyrate may be exploited by cancer cells as a source of energy, which indirectly benefits from tumor growth.^[57] In colorectal cancer, some *E. coli* are found to synthesize a genotoxin called colibactin in human host DNA (double-stranded breaks), which causes genomic instability and drives tumorigenesis.^[58] Similarly, enterotoxigenic *B. fragilis* (ETBF) secretes *B. fragilis* toxin (BFT), which elicits activation of Wnt and NF- κ B pathways, induces hyperproliferation of epithelium and inflammatory responses, thus establishing a microenvironment favorable for tumor growth.^[59] Some bacterial products, such as hydrogen sulfide, ammonia, and nitrate, have a toxic effect on the mucosa and also cause oxidative stress to contribute indirectly to the increased cancer risk.^[60,61] For example, deoxycholic acid, originating from gut bacteria, induces the development of hepatocellular carcinoma by provoking DNA damage and inducing inflammation.^[62] To counterbalance these effects, in some instances, short-chain fatty acids (SCFAs), such as butyrate, propionate, and acetate, have complex, potentially dual effects, both good and bad. Butyrate is generally anticancer in its properties and acts as a histone deacetylase inhibitor to induce apoptosis in cancer cells, inhibit cancer cell proliferation, and promote the activation of immune cells. This environment-dependent variability also means that the effects of SCFAs on the tumor should be considered with reference to the specific tumor type and background metabolism.^[57,63] For example, tumor-associated microbiota-derived butyrate is associated with Wnt/ β -catenin-dependent mechanisms of osteosarcoma metastasis.^[64] Other bacterial metabolites can change the host immune system dramatically. Bacteria can modify dendritic and T-cell differentiation through the secretion of indole metabolites and secondary bile acids, setting up an immunosuppressive environment and weakening host antitumor immune responses. Besides, other studies reveal that specific metabolites have an anticancer activity by promoting effector T-cell activation or stimulating antigen presentation. Such dual function shows the complex biological balance between tumor-associated microbiota and their metabolites within the biological environment.^[62,65,66] Overall, the influence of bacterial metabolites in the TME is neither inherently protumorigenic nor antitumorigenic and is rather dependent

on several aspects, including the type and concentration of the metabolites, host genetic background, and immune status. In-depth investigation of the causal effects of specific metabolites on cancer development not only elucidates the mechanistic connections between bacteria and cancers but may also reveal novel insights for developing precision therapies to regulate metabolites.

2.4.1. Immune regulation of bacteria in tumors. Another important role of bacteria in the TME is the influence on the host's immune system. Under normal physiological conditions, the immune system recognizes and eliminates cells that could become cancerous; but a tumor usually develops in association with immune evasion mechanisms. Some of the recent studies support the idea that intratumoral bacteria can affect the magnitude and inclination of antitumor immune responses in part by directly participating in activation or inhibition of immune cells and, in part, indirectly by downregulating inflammatory signaling mediators and pathways.^[10,30,67] Some bacteria create an immunosuppressive environment for the benefit of the tumor cell and are able to suppress the immune surveillance function of the immune system. For example, *F. nucleatum* plays a role in immune evasion of colorectal cancer, breast cancer, and metastases.^[12,68–70] *F. nucleatum* binds to immune checkpoint receptor protein TIGIT expressed on the surface of human immune cells via the binding protein Fap2 and suppresses the cytotoxic function of natural killer (NK) cells and CD8⁺ T cells, completely short-circuiting the antitumor immune response.^[71] Also, it induces the recruitment and generation of myeloid-derived suppressor cells (MDSCs) and of TAMs that contribute to immune suppression in the tumor environment by secretion of immunosuppressive cytokines, including IL-10 and TGF- β . For example, the bacteria are not only cause immune suppression.^[72,73] Specific circumstances exist in which the tumor-associated microbiota can elicit strong immune responses against tumors and abrogate tumor progression. For some gram-positive bacteria, cell wall peptidoglycans, lipoteichoic acid, and LPS components are recognized by TLRs, which in turn stimulate the action of dendritic cells and macrophages, induce proinflammatory reactions, activate effector T cells, etc.^[74] These immune-stimulating effects can also stimulate the effects of immune checkpoint inhibitors, at least in specific settings. For example, mice studies suggest that the presence of the gut commensal bacterium *Akkermansia muciniphila* strongly improves the efficiency of the PD-1/PD-L1 blockade therapy, revealing the possibilities of adjunct roles of bacteria in cancer immunotherapy.^[75–78] Thus, bacterial effects on immunity are often highly specific and context dependent. The same bacterial species will have very different effects on different hosts or on different types of tumors. For example, SCFAs, important bacterial metabolic products, are capable of promoting expansion of regulatory T cells (via inhibition of histone deacetylases) to promote immune suppression, but can also aid CD8-positive T-cell activity for better antitumor immunity under certain

conditions. Nevertheless, this exchange-reversal behavior further highlights the complexity and highly dynamic interplay of bacteria with the human immune system.^[79,80]

In summary, bacteria present in the TME regulate the immune system via different mechanisms and might facilitate tumor immune evasion while serving as antigen presenting, eliciting antitumor immune responses under certain circumstances. Deeper insights into the mutual, bidirectional regulation between bacteria and the immune system of the host can explain why there are variations in the response to tumor immunotherapy and provide a solid theoretical basis for combination therapy between tumor immunotherapy and modulating treatments of microbiota.

2.4.2. Interactions between bacteria and tumor matrix components. Empirical studies show that the tumor stroma, as part of the tumor milieu, includes the ECM, vascular endothelial cells, CAFs, and various structural and signaling factors. The tumor stroma provides physical and nutrient supply pathways for tumor cells and also exerts a dramatic influence over tumor growth, invasion, and metastasis. Recently, substantial evidence is beginning to accumulate that bacteria engage in highly complicated, layer-of-layer interactions with the tumor stroma components and that they feature prominently in changes in the tumor stroma. Moreover, bacteria remodel the structure and functionality of the ECM by secreting a variety of degradative enzymes. For example, some anaerobic bacteria secrete various enzymes, such as collagenases and hyaluronidases, which speed up the degradation and remodeling of the ECM, thus reducing ECM integrity and promoting tumor cell migration and metastasis. Such ECM disruption not only forms migration opportunities for tumor cells but also provides a favorable environment for bacterial invasion and spread inside the tumor tissue. For example, some strains of bacteria can interfere with the ECM indirectly by influencing the expression of matrix metalloproteinases (MMPs). Several studies demonstrated that contamination of cancer tissues by *H. pylori* is associated with upregulation of MMPs expression, which is a strong mediator of ECM degradation and hence of angiogenesis and tumor metastasis.^[81] This is remarkable and shows that bacteria can indirectly modulate the matrix structure by producing host factors. Interactions between bacteria and CAFs have received increasing attention, as CAFs represent the principal source of ECM production and remodeling, and their activation, secretion properties determine the composition and stiffness of the tumor matrix. For example, bacterial infection can increase secretion by CAFs of collagen and fibronectin to increase matrix stiffening, that is, worsen the malignant phenotype of the tumor cells. However, in response to stimulation by bacteria, CAFs secrete proinflammatory factors such as IL-6 (interleukin-6) and CXCL12 that stimulate proliferation and metastasis in tumor cells but also induce recruitment of immunosuppressive cells into the tumor, thus further modulating the tumor

immune microenvironment.^[82] Whereas bacteria may also stimulate the formation of the matrix vascular network by regulating the vascular endothelial growth factors (VEGFs). Certain bacterial infections can promote tumor angiogenesis by upregulating VEGF; the vascular structure, which may be more disordered and aberrant in the tumor matrix, is generated.^[83–85] Abnormal vascular networks not only supply oxygen and nutrients to tumor cells but also serve as niches for bacterial colonization and excretion of their metabolic products. Such abnormal angiogenesis supports the tumor malignancy and can also impede the therapeutic delivery of antitumor agents.^[86,87] Notably, the action of bacteria on the tumor matrix is not unidirectional, but highly dynamic and bidirectional regulation. In turn, the variations of matrix stiffness, oxygen, and ECM composition regulate the recruitment of bacteria and the bacterial metabolism within the tumor. For example, hypoxic and high-lactate environments not only afford an adequate niche for anaerobic bacteria, but also possibly change the spectrum of their metabolic products and are thus able to further affect the function and signal of matrix cells. These kinds of feedback effects play a critical role in overall increasing the complexity of the TME and adding new viewpoints in the interaction between bacteria and the matrix of tumors.^[88]

Summing up, bacteria secrete enzymes (such as collagenase) that directly degrade the ECM, paving the way for tumor spread. Alterations in ECM structure activate CAFs, which secrete factors like IL-6, further deteriorating the TME. The remodeled matrix in turn provides metabolic niches for bacteria (such as hypoxic zones), creating a vicious cycle.

These effects also contribute to tumor invasion and metastasis and can also affect immune cell infiltration and the contribution of therapies to treatment. Future research of the molecular links between bacteria and matrix should lead to important insights into key steps of tumor progression and to theoretical ideas for treatment exploiting matrix microbiota interactions.

2.5. The influence of bacteria on tumor metastasis

Metastasis of tumors accounts for the overwhelming majority of cancer mortality and encompasses the mechanism in which tumor cells leave the primary tumor, invade the vasculature or lymphatic system, colonize distant sites in secondary organs, and recruit metastatic daughter tumors. Recent research also suggests that bacteria are key players in the TME and, thus, can drive and affect tumor metastases at different stages of initiation and progression. As well as influencing the migratory potential of cancer cells directly by affecting their signal transduction pathways, bacteria facilitate organ metastasis by changing the ECM, modifying immune cell activity and the organ microenvironment.^[89–91] Extensive evidence now suggests that specific bacterial infections can “activate” important signaling pathways in tumor cells and promote their metastatic invasion and migration potential. For example, *E. nucleatum* in

colorectal cancer promotes epithelial–mesenchymal transition (EMT) by mediating β -catenin and NF- κ B signaling pathways, leading to an increased ability of colon tumor cells to become motile and invasive.^[10] Similarly, *H. pylori* infection has been reported to induce EMT in gastric cancer cells and to facilitate their progression and metastasis by perturbing the epithelial cell polarity and lowering cell-to-cell adhesion.^[92–94] For this reason, bacteria cannot be regarded as bystanders in the environment of the tumor but actively participate in producing the invasive phenotype required for metastasis. Besides their direct effect on tumor cells, bacteria can indirectly facilitate tumor metastasis via the host immune response. Some specific bacteria could promote the infiltration of immunosuppressive cells, such as MDSCs, regulatory T cells, and M2-derived macrophages, leading to an immune-suppressive milieu favorable for metastases.^[95,96] Some bacterial infections induce chronic inflammation, and the resulting inflammatory molecules (such as IL-6 and TNF- α) promote angiogenesis and ECM degradation, thereby accelerating the dissemination of tumor cells into the blood and lymphatic circulation.^[97] For example, chronic infections with oral pathogenic bacteria have been reported for metastasis-promoting roles in esophageal cancer and head and neck tumors, with mechanisms presumably working via ongoing activation of the inflammation–immune axis.^[98] More intriguingly, bacteria can also contribute to the generation of a “pre-metastatic niche” at sites distal to the cancer. Certain bacteria can colonize distal organs long before the arrival of tumor cells and precondition distal organs for tumor cell colonization by secretion of metabolic products or modifying local immune responses. For example, experimental evidence suggests that dysbiosis of the gut microbiota disturbs the immune homeostasis of the liver and, therefore, increases survival and proliferation of tumor cells in the liver.^[99–101] Similarly, changes in the microbiome of breast cancer that promote metastatic breast cancer have been related to the development of lung metastasis, suggesting that bacteria influence metastasis in the “seeding and growth” phase by potentially determining the local environment of distant organs.^[102] The bacterial influence on tumor metastasis is at different levels and stages, ranging from direct bacterial effect on tumor cell signals and tumor phenotypes, generating prometastatic TMEs through immune activation and long-term inflammation, to involvement in tissue/organ preconditioning at distant sites. These insights will not only deepen our insight into the complex pathways regulating the metastasis process but also make tumor-associated bacteria attractive targets for therapeutic strategies that aim to restrict metastasis and improve the prognosis for metastatic cancer patients.

3. Clinical relevance and prognostic value of bacteria

During recent years, many clinical studies have demonstrated the importance of tumor-associated bacteria in

cancer initiation and progression and also have developed potential diagnostic and prognostic value. In terms of diagnosis, the presence of a specific bacterium can also be used as a diagnostic marker of early cancer. For example, high *F. nucleatum* levels in tumor tissue, blood, and fecal samples of colorectal cancer patients have all been identified, and they are helpful to conventional screening (e.g., the fecal occult blood test).^[103,104] Such noninvasive screening methods offer new insights into the early screening of high-risk people. Similar to serological testing for *H. pylori* infection, this testing has been widely used for the risk assessment of gastric cancer and surveillance. In terms of prognosis, bacterial abundance in the TME correlated closely with cancer stage, cancer metastasis ability, and patient survival. Several studies have shown an increased abundance of fusobacteria, specifically *F. nucleatum*, in the tumor tissues of colorectal cancer patients and its association with recurrence risk, a shorter progression-free survival (PFS), and shorter overall survival (OS). It was during this same time period that the existence of particular probiotics, especially an enrichment for *Bifidobacterium*, was associated with responses to immunotherapy^[105–107] and may in fact serve as predictors of response.^[108,109] With respect to therapeutic effect, intratumoral bacteria are demonstrated to significantly affect the efficacy of radiotherapy, chemotherapy, and immunotherapy. For example, *F. nucleatum* has been implicated to induce chemoresistance in colorectal cancer cells by inducing an autophagy pathway, whereas some probiotics can enhance the antitumor effects of immune checkpoint inhibitors.^[110–112] The results offer more than merely insights into the predictability of therapeutic outcomes; they also highlight microbial community properties as important references for personalized treatment strategies. Finally, the identification of tumor-associated bacteria provides new possibilities for precision medicine. Analysis of profiles of microbial communities in tumors of patients' cancers provides opportunities for categorizing disease, which implies stratification of patients at risk of disease, thus enabling the development of more individualized treatments. Furthermore, with the advancement of metagenomic sequencing and artificial intelligence (AI), tumor-associated bacteria will soon be put to use in clinical decision systems, ultimately forming important components of precision oncology.

4. Immunotherapy targeting bacteria within tumors

Antimicrobial drugs have been routinely used to manage infection and to prevent complications in cancer patients. However, as has become evident, the role of bacteria in the TME has shifted from the “anti-infection” role that is well defined in conventional chemotherapy to also incorporate the direct manipulation of tumor-associated bacteria and the indirect promotion of antitumor immunity. Overall, current evidence in models of colorectal, breast, and gastric cancer indicates that specific bacterial species,

including *F. nucleatum* and *H. pylori*, mediate tumorigenesis, promote metastasis, and support immune evasion. In this respect, the use of antimicrobial agents to kill or diminish tumor-associated microbiota is a novel adjuvant therapeutic approach, particularly with chemotherapy or immune checkpoint inhibitor therapies, where it could significantly amplify treatment efficacy.^[113–115] Such applications of broad-spectrum antibiotics, however, come with inherent limitations. These agents have no specificity. With these agents eliminating the carcinogenic bacteria, they change the balance of commensal bacteria, predisposing patients to secondary infections and immune disorders. By virtue of its dense ECM, poor vascular perfusion, and hypoxia within solid tumors, drugs cannot readily penetrate and traditional antibiotics do not reach the true centers of solid tumors at a concentration sufficient for their antibacterial activity.^[116,117] Moreover, prolonged use of these agents may result in the emergence of antibiotic resistance and thus loss of the long-term effect of treatment. These shortcomings of classical antibiotics in the treatment of cancers implicate the fundamental insufficiency of conventional antimicrobial agents in this area of application and drive efforts to develop novel therapeutic agents with higher specificity and efficiency in drug targeting. Over the past decade, targeted delivery platforms with the aid of advances in nanotechnology and materials sciences have provided innovative application strategies of antimicrobial drugs. Nanoparticles, thanks to their easy surface modification and their accumulation advantage at the tumor site due to the enhanced permeability and retention effect, have been applied as carriers for antimicrobial agents.^[118] Surface conjugating elements can be added to nanoparticles, bestowing the nanoparticles with affinity to specific bacterial components and thus enabling bacterial selective delivery. An antibody-conjugated liposome with targeting molecules against *F. nucleatum* surface proteins was very potent for both antibacterial and antitumor activity *in vivo* studies.^[119] Moreover, researchers have exploited the special microenvironmental features of tumors—for example, acidic pH, hypoxia, and oxidative stress state (reactive oxygen species, ROS)—to develop different types of stimulus-responsive delivery platforms, and they promote drug release in a tumor-specific activation for cancer-specific accumulation of drugs in the tumor and low off-target toxic effect in normal tissues. Notably, these innovative carriers are not only capable of carrying traditional antibiotics, but can also be mixed with photodynamic therapy or chemotherapeutic agents to induce immunogenic cell death (ICD) of tumor cells. Likewise, the carriers kill bacteria and release damage-associated molecular patterns to further enhance antitumor immunity.^[120,121]

Building on some of these preclinical successes, synergies between antimicrobials and immunotherapy have been demonstrated in diverse preclinical models. Clearance of particular bacteria precludes the blockade of dendritic cells and T cells by bacterial metabolites and, at the same time, increases tumor responsiveness to immune

checkpoint inhibitors. For example, in a mouse model of colorectal cancer, anti-PD-1 blockade efficacy was significantly improved after killing the bacterial strain *F. nucleatum* with metronidazole, showing that antimicrobial treatment might “unlock immunotherapy resistance” that was previously inaccessible.^[122] Similarly, several studies exploiting nanoparticle-based delivery of an antimicrobial have improved microbial burden and shown also improved immune cells’ infiltration, highlighting the link between immunomodulation at the site and immune activation in a tight way.^[123] This dual effect lends support for future integrative interventions on the TME with the aim to elicit synergic elimination of tumor-associated microbial communities and simultaneous boosting of host antitumor immunity. Despite their great promise, antimicrobial agents and antimicrobial targeted delivery vehicles still face challenges in clinical transition. First, the intratumoral microbiome is rather heterogeneous, and there is considerable compositional variability of the intratumoral microbiome across cancer types, even between patients with a given cancer, highlighting the need for personalized antimicrobial treatment regimens. Second, for nanodelivery systems, off-target effects may occur, with “premature” drug release in nontumor tissues potentially altering commensal microbiota or inducing undesired inflammation. Third, bacterial resistance can be alarming, particularly in advanced chronic disease, where the genomic flexibility of bacteria can induce resistance and account for poor long-term therapeutic effects at any stage of tumor growth. Fourth, a significant gap still remains between basic research and clinical research and settings. Almost all the evidence is at the animal model level; the complex and dynamic human microecological environment and its effects on the biodistribution, release, immune modulation, and so on for drugs are inadequate.

Overall, using antimicrobial molecules with antimicrobial delivery agents brings a new opportunity for precise control of tumor microbiota. Given such an opportunity by the simultaneous convergence of nanotechnology, molecular targeting, and immunomodulation, these approaches have huge potential to eliminate protumor microbes and reactivate responses to immunotherapy in each target tissue together. Future work should aim at developing personalized and smart delivery platforms for microbial probiotics appropriate for different types of cancer, along with rigorous clinical testing of their safety and efficacy, to effectively build and revolutionize cancer treatments.

5. Future perspectives

The emerging field of bacteria within the TME offers unprecedented opportunities for cancer treatment, yet its clinical translation still faces numerous challenges requiring future exploration in the following directions.

First, there is a need for more systematic and precise characterization of bacterial lineages and dynamic changes within the TME. Current studies predominantly

rely on static analyses using metagenomic sequencing and tissue sections, which struggle to fully reveal the dynamic roles of bacteria during tumor progression. Future approaches should integrate single-cell omics, spatial transcriptomics, and multimodal imaging technologies to track bacterial distribution, metabolic states, and host–cell interactions within tumor tissues in real time. This will clarify the specific mechanisms of action for different bacteria across cancer types and disease stages, laying the groundwork for personalized therapeutic strategies. Second, immunotherapy targeting tumor-associated bacteria requires further optimization. While current research demonstrates bacteria’s role as key immune regulators, the challenge remains in selectively eliminating tumor-promoting bacteria while preserving or harnessing those with antitumor potential. Future approaches may involve constructing “smart” bacterial regulatory systems through gene editing and synthetic biology to achieve precise microbial reshaping. This could promote immune activation while avoiding excessive inflammation or toxic side effects. Third, the application of engineered bacteria in antitumor therapy must progress toward clinical practice. Most engineered bacterial therapies remain confined to animal model validation, and challenges persist in achieving safe, stable, and controllable human applications. Interdisciplinary collaboration will be the core driver for advancing this field. Research on bacteria within the TME involves not only microbiology, immunology, and oncology but also intersects with materials science, synthetic biology, and AI technologies. Through multidisciplinary integration, it is possible to systematically model tumor–bacteria interaction mechanisms, utilize AI to discover novel microbial biomarkers and potential therapeutic targets, and drive the design and optimization of personalized bacterial treatment regimens. With accumulating clinical trial data and evolving regulatory frameworks, bacteria-based antitumor therapies hold promise for achieving clinical breakthroughs within the next decade.

In summary, research on bacteria within the TME is undergoing rapid development at a critical juncture, demonstrating immense potential in both theoretical and practical applications. From fundamental mechanism exploration to clinical translation, and from harnessing natural bacterial functions to applying engineered modifications, bacteria may emerge as a new pillar of future antitumor therapy. With ongoing advances in research and technology, precision therapy based on bacteria holds promise to overcome the limitations of existing treatments and usher in a new era of cancer therapy.

6. Conclusion

Bacteria are integral to the TME and affect cancer initiation, progression, and therapeutic response. Beyond their classical role as carcinogenic agents, tumor-bacterial microbes induce cell behavior via metabolites and

signaling pathways and reconfigure the immune landscape, influencing tumorigenesis. Therapeutic approaches against these bacteria—such as the selective ablation of protumor species and exploiting the beneficial microbes—may also enhance the effectiveness of immunotherapies. Engineered bacteria are programmed with synthetic biology tools to deliver their effects to tumors, produce therapeutic molecules, and generate immune responses as versatile living therapeutics. There are promising preclinical studies, but clinical translates with hurdles in terms of safety, stability, and regulations. New emerging technologies, including single-cell genomics, spatial transcriptomics, and AI, can help navigate precise bacterial mapping and function spatial characteristics to support biomarker discovery and microbiota targeting studies. Overall, shedding light on the roles of bacteria in tumors offers great insights toward new anti-cancer strategies, putting bacteria into focus in the future of precision oncology.

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Ethical statement

Not applicable.

Conflicts of interest

JD is the Editor-in-Chief of *Bone and Bone Related Cancer Research*, and LJ is the Managing Editor of *Bone and Bone Related Cancer Research*. They were excluded from the peer-review process and all editorial decisions related to the acceptance and publication of this article. Peer-review was handled independently by the other editors to minimize bias. The remaining authors have no conflicts of interest to disclose.

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Data availability statement

The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

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References

- [1] Houthuijzen JM, de Bruijn R, van der Burg E, et al. CD26-negative and CD26-positive tissue-resident fibroblasts contribute to functionally distinct CAF subpopulations in breast cancer. *Nat Commun.* 2023;14:183.
- [2] Labernadie A, Kato T, Brugués A, et al. A mechanically active heterotypic E-cadherin/N-cadherin adhesion enables fibroblasts to drive cancer cell invasion. *Nat Cell Biol.* 2017;19:224–37.
- [3] Sun L, Kees T, Almeida AS, et al. Activating a collaborative innate-adaptive immune response to control metastasis. *Cancer Cell.* 2021;39:1361–74.e9.
- [4] Cheng M, Chen S, Li K, et al. CD276-dependent efferocytosis by tumor-associated macrophages promotes immune evasion in bladder cancer. *Nat Commun.* 2024;15:2818.
- [5] Fu Y, Wang D, Wang H, et al. TSPO deficiency induces mitochondrial dysfunction, leading to hypoxia, angiogenesis, and a growth-promoting metabolic shift toward glycolysis in glioblastoma. *Neuro Oncol.* 2020;22:240–52.
- [6] Végran F, Boidot R, Michiels C, Sonveaux P, Feron O. Lactate influx through the endothelial cell monocarboxylate transporter MCT1 supports an NF- κ B/IL-8 pathway that drives tumor angiogenesis. *Cancer Res.* 2011;71:2550–60.
- [7] Deng F, Zhou R, Lin C, et al. Tumor-secreted dickkopf2 accelerates aerobic glycolysis and promotes angiogenesis in colorectal cancer. *Theranostics.* 2019;9:1001–14.
- [8] Galeano Niño JL, Wu H, LaCourse KD, et al. Effect of the intratumoral microbiota on spatial and cellular heterogeneity in cancer. *Nature.* 2022;611:810–7.
- [9] Nejman D, Livyatan I, Fuks G, et al. The human tumor microbiome is composed of tumor type-specific intracellular bacteria. *Science.* 2020;368:973–80.
- [10] Rubinstein MR, Wang X, Liu W, Hao Y, Cai G, Han YW. *Fusobacterium nucleatum* promotes colorectal carcinogenesis by modulating E-cadherin/ β -catenin signaling via its FadA adhesin. *Cell Host Microbe.* 2013;14:195–206.
- [11] Kostic AD, Chun E, Robertson L, et al. *Fusobacterium nucleatum* potentiates intestinal tumorigenesis and modulates the tumor-immune microenvironment. *Cell Host Microbe.* 2013;14:207–15.
- [12] Parhi L, Alon-Maimon T, Sol A, et al. Breast cancer colonization by *Fusobacterium nucleatum* accelerates tumor growth and metastatic progression. *Nat Commun.* 2020;11:3259.
- [13] Jia D, Wang Q, Qi Y, et al. Microbial metabolite enhances immunotherapy efficacy by modulating T cell stemness in pan-cancer. *Cell.* 2024;187:1651–65.e21.
- [14] Sivan A, Corrales L, Hubert N, et al. Commensal *Bifidobacterium* promotes antitumor immunity and facilitates anti-PD-L1 efficacy. *Science.* 2015;350:1084–9.
- [15] Battaglia TW, Mimpfen IL, Traets JJH, et al. A pan-cancer analysis of the microbiome in metastatic cancer. *Cell.* 2024;187:2324–35.e19.
- [16] Shi H, Chen L, Liu Y, et al. Bacteria-driven tumor microenvironment-sensitive nanoparticles targeting hypoxic regions enhances the chemotherapy outcome of lung cancer. *Int J Nanomedicine.* 2023;18:1299–315.
- [17] Wong-Rolle A, Dong Q, Zhu Y, et al. Spatial meta-transcriptomics reveal associations of intratumor bacteria burden with lung cancer cells showing a distinct oncogenic signature. *J ImmunoTher Cancer.* 2022;10:e004698.
- [18] Chai X, Wang J, Li H, et al. Intratumor microbiome features reveal antitumor potentials of intrahepatic cholangiocarcinoma. *Gut Microbes.* 2023;15:2156255.
- [19] Prudent E, Raoult D. Fluorescence *in situ* hybridization, a complementary molecular tool for the clinical diagnosis of infectious diseases by intracellular and fastidious bacteria. *FEMS Microbiol Rev.* 2019;43:88–107.

- [20] Becerra SC, Roy DC, Sanchez CJ, Christy RJ, Burmeister DM. An optimized staining technique for the detection of Gram positive and Gram negative bacteria within tissue. *BMC Res Notes*. 2016;9:216.
- [21] Yong X, Tang B, Li BS, et al. *Helicobacter pylori* virulence factor CagA promotes tumorigenesis of gastric cancer via multiple signaling pathways. *Cell Commun Signal*. 2015;13:30.
- [22] Qu R, Zhang Y, Ma Y, et al. Role of the gut microbiota and its metabolites in tumorigenesis or development of colorectal cancer. *Adv Sci (Weinh)*. 2023;10:e2205563.
- [23] Little A, Tangney M, Tunney MM, Buckley NE. *Fusobacterium nucleatum*: a novel immune modulator in breast cancer? *Expert Rev Mol Med*. 2023;25:e15.
- [24] Zhao Y, Yang Z, Wu D, Zhao H. Dissecting the intratumoral microbiome landscape in lung cancer. *Front Immunol*. 2025;16:1614731.
- [25] McAllister F, Khan MAW, Helmink B, Wargo JA. The tumor microbiome in pancreatic cancer: bacteria and beyond. *Cancer Cell*. 2019;36:577–9.
- [26] Li C, Chen Y, Yao W, et al. Gut microbiome and serum metabolome alterations in osteosarcoma patients. *Front Microbiol*. 2025;16:1616603.
- [27] Zepeda-Rivera M, Minot SS, Bouzek H, et al. A distinct *Fusobacterium nucleatum* clade dominates the colorectal cancer niche. *Nature*. 2024;628:424–32.
- [28] Bullman S, Pedamallu CS, Sicinska E, et al. Analysis of *Fusobacterium* persistence and antibiotic response in colorectal cancer. *Science*. 2017;358:1443–8.
- [29] Yang Y, Weng W, Peng J, et al. *Fusobacterium nucleatum* increases proliferation of colorectal cancer cells and tumor development in mice by activating toll-like receptor 4 signaling to nuclear factor- κ B, and up-regulating expression of MicroRNA-21. *Gastroenterology*. 2017;152:851–66.e24.
- [30] Rubinstein MR, Baik JE, Lagana SM, et al. *Fusobacterium nucleatum* promotes colorectal cancer by inducing Wnt/ β -catenin modulator annexin A1. *EMBO Rep*. 2019;20:e47638.
- [31] Mondal T, Chattopadhyay D, Saha Mondal P, et al. *Fusobacterium nucleatum* modulates the Wnt/ β -catenin pathway in colorectal cancer development. *Int J Biol Macromol*. 2025;299:140196.
- [32] Li X, Huang J, Yu T, et al. *Fusobacterium nucleatum* promotes the progression of colorectal cancer through Cdk5-activated Wnt/ β -Catenin signaling. *Front Microbiol*. 2020;11:545251.
- [33] Dejea CM, Fathi P, Craig JM, et al. Patients with familial adenomatous polyposis harbor colonic biofilms containing tumorigenic bacteria. *Science*. 2018;359:592–7.
- [34] Joo JE, Chu YL, Georgeson P, et al. Intratumoral presence of the genotoxic gut bacteria pks+ *E. coli*, enterotoxigenic *Bacteroides fragilis*, and *Fusobacterium nucleatum* and their association with clinicopathological and molecular features of colorectal cancer. *Br J Cancer*. 2024;130:728–40.
- [35] Hieken TJ, Chen J, Chen B, et al. The breast tissue microbiome, stroma, immune cells and breast cancer. *Neoplasia*. 2022;27:100786.
- [36] German R, Marino N, Hemmerich C, et al. Exploring breast tissue microbial composition and the association with breast cancer risk factors. *Breast Cancer Res*. 2023;25:82.
- [37] Liu E, Zhang F, Xu T, Ye L, Ma SSQ, Ji ZS. Relationship between tumor microbiota transcriptional activity and gene expression in breast cancer. *BMC Cancer*. 2023;23:252.
- [38] Zhao F, An R, Ma Y, et al. Integrated spatial multi-omics profiling of *Fusobacterium nucleatum* in breast cancer unveils its role in tumour microenvironment modulation and cancer progression. *Clin Transl Med*. 2025;15:e70273.
- [39] Shirai H, Ito C, Tsukada K. pH-taxis drives aerobic bacteria in duodenum to migrate into the pancreas with tumors. *Sci Rep*. 2022;12:1783.
- [40] Riquelme E, Zhang Y, Zhang L, et al. Tumor microbiome diversity and composition influence pancreatic cancer outcomes. *Cell*. 2019;178:795–806.e12.
- [41] Pushalkar S, Hundeyin M, Daley D, et al. The pancreatic cancer microbiome promotes oncogenesis by induction of innate and adaptive immune suppression. *Cancer Discov*. 2018;8:403–16.
- [42] Druzhinin VG, Baranova ED, Demenkov PS, Matskova LV, Larionov AV, Yuzhalin AE. Lower respiratory tract microbiome signatures of health and lung cancer across different smoking statuses. *Cancers (Basel)*. 2025;17:2643.
- [43] Yang J, Mu X, Wang Y, et al. Dysbiosis of the salivary microbiome is associated with non-smoking female lung cancer and correlated with immunocytochemistry markers. *Front Oncol*. 2018;8:520.
- [44] Ying KL, Brasky TM, Freudenheim JL, et al. Saliva and lung microbiome associations with electronic cigarette use and smoking. *Cancer Prev Res (Phila)*. 2022;15:435–46.
- [45] Liu X, Sun W, Ma W, et al. Smoking related environmental microbes affecting the pulmonary microbiome in Chinese population. *Sci Total Environ*. 2022;829:154652.
- [46] Zeng W, Zhao C, Yu M, et al. Alterations of lung microbiota in patients with non-small cell lung cancer. *Bioengineered*. 2022;13:6665–77.
- [47] Wu Z, Tang J, Zhuang R, et al. The microbiome of lower respiratory tract and tumor tissue in lung cancer manifested as radiological ground-glass opacity. *Front Bioeng Biotechnol*. 2022;10:892613.
- [48] Bingula R, Filaire E, Molnar I, et al. Characterisation of microbiota in saliva, bronchoalveolar lavage fluid, non-malignant, peritumoural and tumour tissue in non-small cell lung cancer patients: a cross-sectional clinical trial. *Respir Res*. 2020;21:129.
- [49] Vétizou M, Pitt JM, Daillère R, et al. Anticancer immunotherapy by CTLA-4 blockade relies on the gut microbiota. *Science*. 2015;350:1079–84.
- [50] Zhang C, Wang Y, He M, et al. Mannose enhances immunotherapy efficacy in ovarian cancer by modulating gut microbial metabolites. *Cancer Res*. 2025;85:2468–84.
- [51] Uzelac M, Xin R, Ongkeko WM. Microbiome dysbiosis is associated with castration resistance and cancer stemness in metastatic prostate cancer. *Int J Mol Sci*. 2024;25:3291.
- [52] Le D, Chambers MM, Mercado K, Gutowski CJ. Characterization of the gut microbiome in an osteosarcoma mouse model. *J Orthop Res*. 2023;41:2730–9.
- [53] Dutta RK, Abu YF, Tao J, et al. Altered gut microbiome drives heightened pain sensitivity in a murine model of metastatic triple-negative breast cancer. *Am J Cancer Res*. 2024;14:274–99.
- [54] Ghaddar B, Biswas A, Harris C, et al. Tumor microbiome links cellular programs and immunity in pancreatic cancer. *Cancer Cell*. 2022;40:1240–53.e5.
- [55] Zhu X, Hu M, Huang X, et al. Interplay between gut microbial communities and metabolites modulates pan-cancer immunotherapy responses. *Cell Metab*. 2025;37:806–23.e6.
- [56] Ujlaki G, Kovács T, Vida A, et al. Identification of bacterial metabolites modulating breast cancer cell proliferation and epithelial-mesenchymal transition. *Molecules*. 2023;28:5898.
- [57] He Y, Fu L, Li Y, et al. Gut microbial metabolites facilitate anti-cancer therapy efficacy by modulating cytotoxic CD8+ T cell immunity. *Cell Metab*. 2021;33:988–1000.e7.
- [58] Pleguezuelos-Manzano C, Puschhof J, Rosendahl Huber A, et al.; Genomics England Research Consortium. Mutational signature in colorectal cancer caused by genotoxic pks+ *E. coli*. *Nature*. 2020;580:269–73.
- [59] Ghobashi AH, Lanzloth R, Ladaika CA, Masood A, O'Hagan HM. Single-cell profiling reveals the impact of genetic alterations on the differentiation of inflammation-induced murine colon tumors. *Cancers (Basel)*. 2024;16:2040.

- [60] Roje B, Zhang B, Mastorilli E, et al. Gut microbiota carcinogen metabolism causes distal tissue tumours. *Nature*. 2024;632:1137–44.
- [61] Kawahara Y, Hirashita Y, Tamura C, et al. *Helicobacter pylori* infection modulates endogenous hydrogen sulfide production in gastric cancer AGS cells. *Helicobacter*. 2020;25:e12732.
- [62] Liu H, Qin CY, Han GQ, Xu HW, Meng M, Yang Z. Mechanism of apoptotic effects induced selectively by ursodeoxycholic acid on human hepatoma cell lines. *World J Gastroenterol*. 2007;13:1652–8.
- [63] Arpaia N, Campbell C, Fan X, et al. Metabolites produced by commensal bacteria promote peripheral regulatory T-cell generation. *Nature*. 2013;504:451–5.
- [64] Yu J, Lu W, Chen Y, et al. Butyrate derived from intratumoral microbiome facilitates pulmonary metastasis in osteosarcoma. *Pharmacol Res*. 2025;219:107897.
- [65] Varanasi SK, Chen D, Liu Y, et al. Bile acid synthesis impedes tumor-specific T cell responses during liver cancer. *Science*. 2025;387:192–201.
- [66] Shen R, Ke L, Li Q, et al. Abnormal bile acid-microbiota cross-talk promotes the development of hepatocellular carcinoma. *Hepatol Int*. 2022;16:396–411.
- [67] Lam KC, Araya RE, Huang A, et al. Microbiota triggers STING-type I IFN-dependent monocyte reprogramming of the tumor microenvironment. *Cell*. 2021;184:5338–56.e21.
- [68] Xu C, Fan L, Lin Y, et al. *Fusobacterium nucleatum* promotes colorectal cancer metastasis through miR-1322/CCL20 axis and M2 polarization. *Gut Microbes*. 2021;13:1980347.
- [69] Chen G, Gao C, Jiang S, et al. *Fusobacterium nucleatum* outer membrane vesicles activate autophagy to promote oral cancer metastasis. *J Adv Res*. 2024;56:167–79.
- [70] Chen S, Su T, Zhang Y, et al. *Fusobacterium nucleatum* promotes colorectal cancer metastasis by modulating KRT7-AS/KRT7. *Gut Microbes*. 2020;11:511–25.
- [71] Gur C, Ibrahim Y, Isaacson B, et al. Binding of the Fap2 protein of *Fusobacterium nucleatum* to human inhibitory receptor TIGIT protects tumors from immune cell attack. *Immunity*. 2015;42:344–55.
- [72] Sakamoto Y, Mima K, Ishimoto T, et al. Relationship between *Fusobacterium nucleatum* and antitumor immunity in colorectal cancer liver metastasis. *Cancer Sci*. 2021;112:4470–7.
- [73] Liang M, Liu Y, Zhang Z, et al. *Fusobacterium nucleatum* induces MDSCs enrichment via activation the NLRP3 inflammasome in ESCC cells, leading to cisplatin resistance. *Ann Med*. 2022;54:989–1003.
- [74] Altves S, Guclu E, Yetisgin E, Bilecen K, Vural H. Upregulation of immune checkpoint PD-L1 in colon cancer cell lines and activation of T cells by *Leuconostoc mesenteroides*. *World J Microbiol Biotechnol*. 2024;40:204.
- [75] Derosa L, Routy B, Thomas AM, et al. Intestinal *Akkermansia muciniphila* predicts clinical response to PD-1 blockade in patients with advanced non-small-cell lung cancer. *Nat Med*. 2022;28:315–24.
- [76] Xu Y, Tan X, Yang Q, Fang Z, Chen W. *Akkermansia muciniphila* outer membrane protein regulates recruitment of CD8⁺ T cells in lung adenocarcinoma and through JAK-STAT signaling pathway. *Microb Biotechnol*. 2024;17:e14522.
- [77] Zhu Z, Huang J, Zhang Y, et al. Landscape of tumoral ecosystem for enhanced anti-PD-1 immunotherapy by gut *Akkermansia muciniphila*. *Cell Rep*. 2024;43:114306.
- [78] Zhu J, Qin S, Gu R, Ji S, Wu G, Gu K. Amuc_1434 from *Akkermansia muciniphila* enhances CD8⁺ T cell-mediated anti-tumor immunity by suppressing PD-L1 in colorectal cancer. *FASEB J*. 2025;39:e70540.
- [79] Yang W, Yu T, Huang X, et al. Intestinal microbiota-derived short-chain fatty acids regulation of immune cell IL-22 production and gut immunity. *Nat Commun*. 2020;11:4457.
- [80] Luu M, Riester Z, Baldrich A, et al. Microbial short-chain fatty acids modulate CD8⁺ T cell responses and improve adoptive immunotherapy for cancer. *Nat Commun*. 2021;12:4077.
- [81] Karayiannis I, Martinez-Gonzalez B, Kontizas E, et al. Induction of MMP-3 and MMP-9 expression during *Helicobacter pylori* infection via MAPK signaling pathways. *Helicobacter*. 2023;28:e12987.
- [82] Kobayashi H, Gieniec KA, Lannagan TRM, et al. The origin and contribution of cancer-associated fibroblasts in colorectal carcinogenesis. *Gastroenterology*. 2022;162:890–906.
- [83] Andriessen EM, Wilson AM, Mawambo G, et al. Gut microbiota influences pathological angiogenesis in obesity-driven choroidal neovascularization. *EMBO Mol Med*. 2016;8:1366–79.
- [84] Cane G, Ginouvès A, Marchetti S, et al. HIF-1 α mediates the induction of IL-8 and VEGF expression on infection with Afa/Dr diffusely adhering *E. coli* and promotes EMT-like behaviour. *Cell Microbiol*. 2010;12:640–53.
- [85] Mendes RT, Nguyen D, Stephens D, et al. Endothelial cell response to *Fusobacterium nucleatum*. *Infect Immun*. 2016;84:2141–8.
- [86] Stylianopoulos T, Jain RK. Combining two strategies to improve perfusion and drug delivery in solid tumors. *Proc Natl Acad Sci U S A*. 2013;110:18632–7.
- [87] Hamzah J, Jugold M, Kiessling F, et al. Vascular normalization in Rgs5-deficient tumours promotes immune destruction. *Nature*. 2008;453:410–4.
- [88] Jalili-Firoozinezhad S, Gazzaniga FS, Calamari EL, et al. A complex human gut microbiome cultured in an anaerobic intestine-on-a-chip. *Nat Biomed Eng*. 2019;3:520–31.
- [89] Fu A, Yao B, Dong T, et al. Tumor-resident intracellular microbiota promotes metastatic colonization in breast cancer. *Cell*. 2022;185:1356–72.e26.
- [90] Bertocchi A, Carloni S, Ravenda PS, et al. Gut vascular barrier impairment leads to intestinal bacteria dissemination and colorectal cancer metastasis to liver. *Cancer Cell*. 2021;39:708–24.e11.
- [91] Zhu Z, Huang J, Li X, et al. Gut microbiota regulate tumor metastasis via circRNA/miRNA networks. *Gut Microbes*. 2020;12:1788891.
- [92] Fang Z, Zhang W, Wang H, et al. *Helicobacter pylori* promotes gastric cancer progression by activating the TGF- β /Smad2/EMT pathway through HKDC1. *Cell Mol Life Sci*. 2024;81:453.
- [93] Chang H, Kim N, Park JH, et al. *Helicobacter pylori* might induce TGF- β 1-mediated EMT by means of cagE. *Helicobacter*. 2015;20:438–48.
- [94] Lee SD, Jeong H, Hwang BR, et al. *Helicobacter pylori* promotes epithelial-to-mesenchymal transition by downregulating CK2 β in gastric cancer cells. *Biochim Biophys Acta Mol Basis Dis*. 2023;1869:166588.
- [95] Zhang Q, Ma C, Duan Y, et al. Gut microbiome directs hepatocytes to recruit MDSCs and promote cholangiocarcinoma. *Cancer Discov*. 2021;11:1248–67.
- [96] Liu JL, Xu X, Rixiati Y, et al. Dysfunctional circadian clock accelerates cancer metastasis by intestinal microbiota triggering accumulation of myeloid-derived suppressor cells. *Cell Metab*. 2024;36:1320–34.e9.
- [97] Larsson BM, Larsson K, Malmberg P, Palmberg L. Gram positive bacteria induce IL-6 and IL-8 production in human alveolar macrophages and epithelial cells. *Inflammation*. 1999;23:217–30.
- [98] Rosa DD, Pasqualotto AC, Denning DW. Chronic mucocutaneous candidiasis and oesophageal cancer. *Med Mycol*. 2008;46:85–91.
- [99] Cheng P, Wu J, Zong G, et al. Capsaicin shapes gut microbiota and pre-metastatic niche to facilitate cancer metastasis to liver. *Pharmacol Res*. 2023;188:106643.

- [100] Yan L, Tan Q, Zhu J, He Y, Tao P, He J. Gut microbiota and glycochenodeoxycholic role in liver metastasis of breast cancer. *Biochem Pharmacol.* 2025;241:117128.
- [101] Yuan N, Li X, Wang M, et al. Gut microbiota alteration influences colorectal cancer metastasis to the liver by remodeling the liver immune microenvironment. *Gut Liver.* 2022;16:575–88.
- [102] Ma T, Tang Y, Wang T, et al. Chronic pulmonary bacterial infection facilitates breast cancer lung metastasis by recruiting tumor-promoting MHCIIhi neutrophils. *Signal Transduct Target Ther.* 2023;8:296.
- [103] Xing C, Zhihao L, Ji D. Diagnostic value of fecal *Fusobacterium nucleatum* in colorectal cancer. *Arch Med Sci.* 2023;19:1929–33.
- [104] Kurt M, Yumuk Z. Diagnostic accuracy of *Fusobacterium nucleatum* IgA and IgG ELISA test in colorectal cancer. *Sci Rep.* 2021;11:1608.
- [105] Xie Y, Jiao X, Zeng M, et al. Clinical significance of *Fusobacterium nucleatum* and microsatellite instability in evaluating colorectal cancer prognosis. *Cancer Manag Res.* 2022;14:3021–36.
- [106] Lee DW, Han SW, Kang JK, et al. Association between *Fusobacterium nucleatum*, pathway mutation, and patient prognosis in colorectal cancer. *Ann Surg Oncol.* 2018;25:3389–95.
- [107] Mima K, Nishihara R, Qian ZR, et al. *Fusobacterium nucleatum* in colorectal carcinoma tissue and patient prognosis. *Gut.* 2016;65:1973–80.
- [108] Mager LF, Burkhard R, Pett N, et al. Microbiome-derived inosine modulates response to checkpoint inhibitor immunotherapy. *Science.* 2020;369:1481–9.
- [109] Zhao H, Li D, Liu J, et al. *Bifidobacterium breve* predicts the efficacy of anti-PD-1 immunotherapy combined with chemotherapy in Chinese NSCLC patients. *Cancer Med.* 2023;12:6325–36.
- [110] Dong J, Li Y, Xiao H, et al. Oral microbiota affects the efficacy and prognosis of radiotherapy for colorectal cancer in mouse models. *Cell Rep.* 2021;37:109886.
- [111] Benej M, Hoyd R, Kreamer M, et al.; exORIEN Consortium. The tumor microbiome reacts to hypoxia and can influence response to radiation treatment in colorectal cancer. *Cancer Res Commun.* 2024;4:1690–701.
- [112] Yu T, Guo F, Yu Y, et al. *Fusobacterium nucleatum* promotes chemoresistance to colorectal cancer by modulating autophagy. *Cell.* 2017;170:548–63.e16.
- [113] Wang J, Deng R, Chen S, et al. *Helicobacter pylori* CagA promotes immune evasion of gastric cancer by upregulating PD-L1 level in exosomes. *iScience.* 2023;26:108414.
- [114] Liu S, Zhang N, Ji X, Yang S, Zhao Z, Li P. *Helicobacter pylori* CagA promotes gastric cancer immune escape by upregulating SQLE. *Cell Death Dis.* 2025;16:17.
- [115] Zhang T, Li Y, Zhai E, et al. Intratumoral *Fusobacterium nucleatum* recruits tumor-associated neutrophils to promote gastric cancer progression and immune evasion. *Cancer Res.* 2025;85:1819–41.
- [116] Jain RK, Baxter LT. Mechanisms of heterogeneous distribution of monoclonal antibodies and other macromolecules in tumors: significance of elevated interstitial pressure. *Cancer Res.* 1988;48(24 Pt 1):7022–32.
- [117] Netti PA, Berk DA, Swartz MA, Grodzinsky AJ, Jain RK. Role of extracellular matrix assembly in interstitial transport in solid tumors. *Cancer Res.* 2000;60:2497–503.
- [118] Shi Y, van der Meel R, Chen X, Lammers T. The EPR effect and beyond: strategies to improve tumor targeting and cancer nanomedicine treatment efficacy. *Theranostics.* 2020;10:7921–4.
- [119] Chen L, Zhao R, Shen J, et al. Antibacterial *Fusobacterium nucleatum*-mimicking nanomedicine to selectively eliminate tumor-colonized bacteria and enhance immunotherapy against colorectal cancer. *Adv Mater.* 2023;35:e2306281.
- [120] Liu X, Sun M, Pu F, Ren J, Qu X. Transforming intratumor bacteria into immunopotentiators to reverse cold tumors for enhanced immuno-chemodynamic therapy of triple-negative breast cancer. *J Am Chem Soc.* 2023;145:26296–307.
- [121] Geng S, Guo P, Li X, et al. Biomimetic nanovehicle-enabled targeted depletion of intratumoral *Fusobacterium nucleatum* synergizes with PD-L1 blockade against breast cancer. *ACS Nano.* 2024;18:8971–87.
- [122] Jiang SS, Xie YL, Xiao XY, et al. *Fusobacterium nucleatum*-derived succinic acid induces tumor resistance to immunotherapy in colorectal cancer. *Cell Host Microbe.* 2023;31:781–97.e9.
- [123] Lu Q, Kou D, Lou S, et al. Nanoparticles in tumor microenvironment remodeling and cancer immunotherapy. *J Hematol Oncol.* 2024;17:16.