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Intratumor microbiota: a new perspective in cancer initiation, development, and therapy

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Abstract

Microbes have been identified as significantly impacting human health. Considerable attention has been focused on how microbiota affects cancer initiation, development, and therapeutic response. Currently, the biological functions of intratumor microbiota have been preliminarily elucidated in the tumors with high microbial abundance. However, the biological roles of the microbiota and their clinical significance in the tumors with low microbial abundance, to our knowledge, remain largely unexplored. This gap in understanding is primarily due to the limited sensitivity of current detection technologies. This review provides a detailed examination of intratumor microbiota characteristics and their interactions with the tumor microenvironment, focusing on microbiota composition in various systems and its clinical role in different tumor types. Furthermore, the review explores the potential applications of intratumor microbiota in cancer immunotherapy, including their role as immune enhancers, new drug delivery targets, and anticancer therapeutic agents. In conclusion, these insights may facilitate the use of microbiota for cancer diagnosis, prognosis, and the development of new therapeutic strategies.

Keywords: Intratumor microbiota, tumor microenvironment, immunotherapy, drug delivery

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1. Introduction

Cancer is a significant global health issue (1). Tumor therapy initially targets molecules like genes, DNA, and proteins, then proceeds to precise subcellular organelles, including mitochondria, endoplasmic reticulum, lysosomes, and nuclei. Mitochondria likely originated from an ancient endosymbiotic event wherein proteobacteria were engulfed to facilitate energy production and cellular metabolism(2), which implies bacteria can integrate into host cells and fulfill specific biological functions. The human microbiome consists of bacteria, fungi, viruses, and archaea. These organisms are abundant in the digestive tract and also present in smaller numbers in the skin, upper respiratory tract, eye, and urogenital tract, which play a key role in various tumors. To date, considerable research has focused on the effect of non-resident bacteria on cancer development and therapeutic response.

Recent advancements in detection methodologies, particularly next-generation sequencing for the analysis of trace DNA, have significantly improved the precision of bacterial detection within tumors and enabled the identification of specific bacterial species. These technological developments challenge the previously held assumption that the presence of bacteria was merely a consequence of contamination during detection processes. Intratumor microbiota may either promote or inhibit cancer initiation, progression, and response to immunotherapy. The role of intratumor microbiota in tumorigenesis depends on its composition and abundance, tumor stage, and the host's immune response. Thus, understanding the microbiota composition across various tumors and their role in cancer initiation and progression can aid in identifying new therapeutic strategies and targets, enhancing treatment efficacy.

In this review, we discuss the characteristics and composition of intratumor microbiota, their critical role in various tumor tissues, and potential applications in cancer immunotherapy. We emphasize the progress of intratumor microbiota across various tumors. This work identifies the microbiota as a tool for cancer diagnosis or prognosis, as well as a new therapeutic strategy.

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2. Characteristics of Intratumor Microbiota in Tumor Tissue

2.1 Diverse sources

Intratumor microbiota can originate from three sources: (i) the primary tumor site, where the microorganisms reside in the tissue that gives rise to the tumor. For instance, Porphyromonas gingivalis in the oral cavity can promote oral squamous cell carcinoma progression(3), and Helicobacter pylori in the stomach can promote gastric cancer development(4); (ii) Normal adjacent tissues (NATs), from which microbiota can migrate into tumor tissues. Nejman et al. found that the bacterial composition of tumor tissues closely resembles that of NATs(5); (iii) Circulation, through which intratumor microbiota, primarily found in tumor and immune cells, can migrate to distant tumor tissues via the bloodstream. Intestinal bacteria can reach different tumor sites via the different organ-gut axis.

2.2 High heterogenicity

Intratumor microbiota compositions exhibit variation across different tumors. Analyses of the Cancer Genome Atlas (TCGA) database, including genome-wide and transcriptome-wide approaches, reveal characteristics of intratumor microbiota. Nejman et al. demonstrated microbial composition variation in different tumors by analyzing more than 1,500 patient tumor samples(5). Bullman et al. used 16S rDNA sequencing on 44 tumor tissues isolated from 11 patients with colorectal cancer (CRC) to reveal varying degrees of heterogeneity in CRC's intratumor microbiota(6). Further, microorganisms were confirmed to exhibit heterogeneous spatial distribution. In addition, intratumor microbiota composition may even vary among different subtypes of the same tumor type(5). Intratumor microbiota is primarily bacterial, but similar characteristics are observed in intratumor fungi. The diversity and abundance of cancer-type-specific fungi are generally lower than those of the corresponding bacterial populations(7). Notably, bacterial and fungal abundances, diversities, and co-occurrences are strongly positively correlated in several tumors. Tumor microenvironment(TME) may provide non-competitive spaces for microbial

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colonization. This contrasts with the gut, where bacterial and fungal populations compete for resources, especially under antitumor or antibiotic therapies.

2.3 Spatiotemporal Dynamics of the Intratumor Microbiome

The intratumoral microbiota demonstrates spatiotemporal dynamics, characterized by systematic reprogramming of its diversity, abundance, and functionality in response to tumor progression, therapeutic interventions, and alterations in the host microenvironment. Temporally, the lung cancer microbiome undergoes evolution with advancing disease stages: early-stage tumors exhibit greater microbial diversity with a predominance of Actinobacteria, whereas late-stage lesions are enriched with butyrate-producing bacteria, such as Roseburia spp. Microbial-derived butyrate functions by inhibiting histone deacetylase HDAC2, enhancing H3K27 acetylation at the promoter region of the long non-coding RNA H19, and inducing M2 macrophage polarization, collectively facilitating metastatic progression(8) Spatially, microbiota establish heterogeneous microniches within tumors. For example, in oral squamous cell carcinoma and colorectal cancer, bacterial communities colonize hypovascularized and highly immunosuppressive microenvironments associated with malignant cells(6). Therapeutically, interventions significantly alter the composition of the microbiome: immune checkpoint blockade (ICB) decreases microbial richness across various tumor models(9). In contrast, shifts in intratumoral microbiota play a critical role in modulating the efficacy of ICB. For instance, the translocation of the probiotic Limosilactobacillus reuteri to melanoma sites enhances the response to ICB through the production of indole-3-aldehyde (I3A), a catabolite of tryptophan(10). In conclusion, the dynamic evolution and biological functions of intratumoral microbiota are governed by a tripartite regulation involving stage progression, therapy-induced perturbations, and metabolic feedback. Their spatiotemporal heterogeneity may offer new insights for prognostic stratification and the development of targeted interventions.

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3. Progress of the intratumor microbiota in various types of tumors

3.1 Digestive system neoplasm

3.1.1 Esophageal cancer

Esophageal cancer (ESCA) is divided into two main histological subtypes: esophageal squamous cell carcinoma (ESCC) and esophageal adenocarcinoma(11). Microbes such as *Fusobacteria*, *Lactobacillales*, *Clostridia*, *Proteobacteria*, and *Negativicutes* are correlated with the clinical characteristics of patients with ESCA (12). Intratumor microbiota enhances antitumor immunity by recruiting the infiltration of immune cells. *Streptococcus* enrichment is often linked to increased CD8+ T cell infiltration and a favorable response to anti-PD-1 therapy(13). In addition, *Fusobacterium*, identified as a pathogen, accelerates ESCC tumorigenesis and metastasis via inducing DNA damage, recruiting myeloid-derived suppressor cells (MDSCs)(14), activating the NF-κβ pathway(15), and increasing METTL3-mediated m6A methylation(16). *Fusobacterium nucleatum* invades senescent ESCC cells, enhancing senescence-associated secretory phenotype secretion, and thereby promoting ESCC progression(17). Zhang et al. reported that *Fusobacterium nucleatum* inhibits T cell proliferation and cytokine secretion, attenuating antitumor immunity in ESCC(18). These findings highlight the potential for intratumoral microbes and their associated metabolites to influence the tumor immune microenvironment and the efficacy of immunotherapy.

3.1.2 Gastric cancer

The gastric cancer microbiota is characterized by reduced diversity and enriched *Oceanobacter*, *Methylobacterium*, and *Syntrophomonas* genera. *Helicobacter pylori*, a common inhabitant, is classified as a class I carcinogen by the WHO (19). However, *Helicobacter pylori* is not the only microbe accelerating gastric cancer progression. Yu et al. identified *Streptococcus anginosus* as a gastric tumorigenesis-promoting pathogen (Figure 1A)(20). Li et al. have demonstrated that the *Streptococcus anginosus* group

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consistently upregulates all metabolites associated with arginine metabolism in gastric cancer tumor specimens. This metabolic reprogramming facilitates tumor cell proliferation, migration, and invasion, while simultaneously inhibiting the differentiation and infiltration of $CD8^+$ T lymphocytes within the tumor immune microenvironment(TIME). These processes collectively contribute to the tumorigenesis and progression of gastric cancer(21).

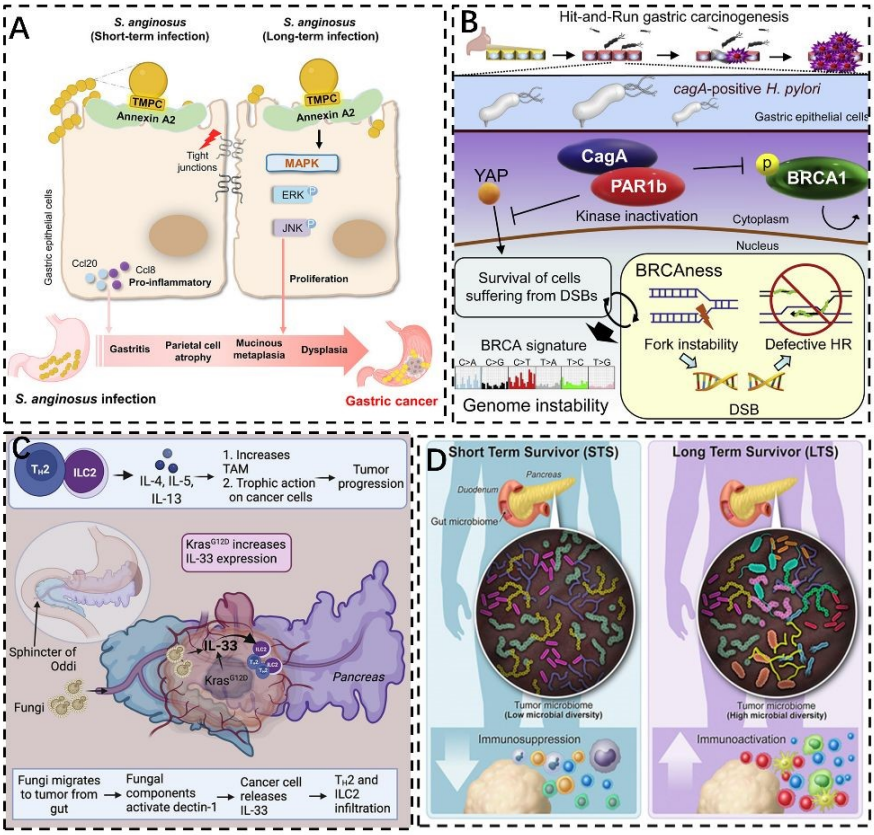

Figure 1. (**A)** Schematic illustration of *Streptococcus anginosus* promotes gastric inflammation, atrophy, and tumorigenesis in mice. Reproduced with permission(20). Copyright, 2024, Elsevier Inc. (**B)** Schematic depiction of *Helicobacter pylori* CagA elicits BRCAness to induce genome instability. Reproduced with permission (25). Copyright, 2021, Elsevier Inc. (**C)** Schematic depiction of fungal mycobiome drives IL-33 secretion and type 2 immunity in pancreatic cancer. Reproduced with permission(52). Copyright, 2022, Elsevier Inc. **(D)** Schematic depiction of PDAC LTS display high tumor microbial diversity and immunoactivation. Reproduced with

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Helicobacter pylori promotes gastric tumorigenesis via mechanisms such as DNA damage(22), oncogenic pathway activation (23), and induction of chronic inflammation and apoptosis(24). Specific virulence factors, including the cytotoxin-associated gene A (CagA) and vacuolating cytotoxin A (VacA), are critical in inducing host cell DNA damage. CagA induces DNA double-strand breaks and disrupts error-free DNA repair via homologous recombination, contributing to gastric carcinogenesis (Figure 1B)(25). VacA induces vacuolization, necrosis, and apoptosis(26). The link between gastric cancer and *Helicobacter pylori* is one of the strongest between a single bacterium and cancer causation. Prospective data indicate that *Helicobacter pylori* precedes tumorigenesis, and antibiotic eradication may reduce gastric cancer incidence.

3.1.3 Colorectal cancer

Gut microbiota, found in CRC, engages in structured crosstalk with the host, influencing multiple physiological processes. CRC-promoting bacterial species like *Fusobacterium nucleatum, Escherichia coli,* and *Bacteroides fragilis*, and CRC-protecting bacterial species like *Clostridium butyricum, Streptococcus thermophilus,* and *Lacticaseibacillus paracasei* are part of the gut microbiota and play critical roles in the development of CRC(27).

Intratumor microbiota produces genotoxins that damage colonic epithelial cell DNA, promoting CRC development. *Escherichia coli* harboring polyketide synthetase pathogenicity island, known to encode the genotoxin colibactin, are increasingly associated with CRC(28-30). *Escherichia coli* that produce colibactin induce DNA breaks, cell cycle arrest, and senescence, promoting tumor growth(31). *Campylobacter jejuni* produces a cytolethal-distending toxin that causes DNA double-strand breaks(32). Moreover, the toxin produced by enterotoxigenic *Bacteroides fragilis* compromises colonic epithelial and barrier integrity, causing inflammation and inducing epithelial cell proliferation through activating the NF-κβ and STAT3 signaling pathways,

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promoting CRC progression(33). Recent evidence suggests that microbiota may drive tumor metastasis by regulating CRC metabolism. *Fusobacterium nucleatum,* a key periodontal pathogen, is enriched in CRC. *Fusobacterium nucleatum* subspecies animalis (Fna) exists in two distinct clades: Fna C1 and Fna C2, with only the latter shown to induce tumors and promote oxidative stress in intestinal metabolism in mouse models(34). *Fusobacterium nucleatum* activates the TLR4/Keap1/NRF2 pathway, increasing CYP2J2 and 12,13-EpOME levels, which promote tumor metastasis(35). Chu et al. reported that trans-3-indoleacrylic acid, a tryptophan metabolite from *Peptostreptococcus anaerobius*, promotes colorectal carcinogenesis via inhibiting ferroptosis (36). These studies indicate that gut microbes can indirectly affect the tumor microenvironment through metabolites or the immune system, potentially altering the composition and function of the intratumoral microbiota.

3.1.4 Liver cancer

Primary liver cancer is classified into three main histological subtypes: hepatocellular carcinoma (HCC), intrahepatic cholangiocarcinoma (ICC), and the combined HCC-ICC(37). HCC is the most prevalent type of liver cancer, characterized by high recurrence rates and poor prognosis(38). Microbial diversity in HCC tissues significantly exceeds that in adjacent tissues. *Bacteroidetes*, *Firmicutes,* and *Proteobacteria* are more abundant in HCC tissues, whereas antitumor bacteria like *Pseudomonas* are less so (39). Cai et al. identified *Burkholderiales*, *Pseudomonadales*, *Xanthomonadales*, *Bacillales,* and *Clostridiales* as the predominant bacterial orders in ICC. An increased presence of *Paraburkholderia fungorum* in paracancerous tissues suggests a role in ICC pathogenesis(40).

Microorganisms contribute to HCC development via direct and indirect mechanisms. (41). *Stenotrophomonas maltophilia* is found in HCC patients with cirrhosis. *Stenotrophomonas maltophilia* activates the TLR4/NF-κβ/NLRP3 pathway, resulting in a senescence-associated secretory phenotype in hepatic stellate cells, which induces inflammation and promotes cirrhosis and HCC progression(42). Mao et al.

found that CD68$^+$ macrophages were more prevalent in areas rich in intratumor microbiota(43). Comparative analysis of HCC subtypes shows that the bacteria-dominant subtype increased M2 macrophage infiltration and upregulated metabolic pathways compared to the virus-dominated subtype. This infiltration of M2 macrophages correlates positively with amino acid metabolism. Notably, HCC metabolic patterns differ significantly from those in normal liver tissues, indicating that the unique microbial environments in HCC patients may influence metabolic variations(44). These findings suggest that the intratumor microbiota could modulate the tumor immune microenvironment through metabolic reprogramming.

3.1.5 Pancreatic cancer

The pancreatic cancer microbiome, communicating with the gut microbiome, affects the host immune response and the disease's natural history. The predominant bacterial genera in the pancreas are *Proteobacteria*, *Bacteroidetes*, and *Firmicutes*. *Enterobacteriaceae* and *Pseudomonadaceae*, identified in pancreatic ductal adenocarcinoma (PDAC), may modulate tumor sensitivity to gemcitabine (Gem)(45).

Intratumor microbiota can remodel the tumor immune microenvironment, accelerating pancreatic cancer development. Bacteria linked to PDAC trigger innate and adaptive immune suppression, reducing MDSCs amount and enhancing M1 macrophage and TH1 differentiation, thereby activating CD8$^+$ T cells (46). Indole-producing bacteria, including *Lactobacillus murinus*, elevate aryl hydrocarbon receptor transcriptional responses and promote an immunosuppressive TME in PDAC, promoting tumor growth(47). Moreover, colonization within the microenvironment of PDAC results in the metabolic production of butyrate. This meta *Clostridium butyricum* bolite increases the susceptibility of PDAC tumor cells to ferroptosis inducers, such as RSL3. The combined therapeutic approach, utilizing either *Clostridium butyricum* colonization or exogenous butyrate supplementation alongside RSL3, synergistically amplifies the inhibitory impact on the proliferation of PDAC tumor cells(48). *Porphyromonas gingivalis*, a key periodontal pathogen, is strongly associated with

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pancreatic cancer(49). Ma et al. reported that *Porphyromonas gingivalis* boosts neutrophil chemokines and elastase secretion, creating a proinflammatory TME and promoting pancreatic cancer progression(50). Nussbaum et al. found that *Porphyromonas gingivalis* protects tumor cells from ROS-induced cell death due to nutrient stress, accelerating PDAC progression(51). Intratumor fungi stimulate IL-33 secretion and type 2 immunity, potentially promoting tumor growth (Figure 1C)(52). *Malassezia* activates mannose-binding lectin, initiating the complement cascade and promoting PDAC progression(53). However, the intratumor microbiota in pancreatic cancer may not be invariably harmful, with some microbes linked to better clinical outcomes. Intratumor microbiota may modulate immune infiltration in pancreatic cancer long-term survivors, thereby inhibiting tumor progression. Riquelme et al. demonstrate that gut bacteria from long-term survival patients modulate pancreatic intratumor bacterial composition, enhance tumor $CD8^+$ T cell activation, and inhibit MDSCs and regulatory T cells (Tregs) accumulation, thereby inhibiting tumor growth (Figure 1D)(54). Furthermore, De et al. found that a subset of tumors contains somatic-cell-associated bacteria, primarily associated with tumor cells and rarely found in nonmalignant tissues(55). This suggests that microbiome targeting may prevent oncogenesis, reverse intratumoral immune tolerance, and enhance the effectiveness of checkpoint-based immunotherapy.

3.2 Respiratory system neoplasms

3.2.1 Lung cancer

Microorganisms are closely associated with lung cancer(56), and the predominant bacteria in lung cancer are *Proteobacteria* and *Actinobacteria*(5). Fungi have been identified within lung cancer cells. Smokers with lung cancer have more abundant intratumor fungi, including higher levels of *Aspergillus* and *Umbelliferae fungi*. Intratumor fungi may serve as markers to effectively distinguish lung cancer from healthy controls, holding potential for early diagnosis. In addition, Goto et al. suggest that the *John Cunningham virus* may contribute to lung cancer progression(57).

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Intratumor microbiota in lung cancer can modulate cytokine production and foster a chronic inflammatory microenvironment that promotes tumorigenesis. Tyler et al. reported that lung microbiota activates γδ T cells, triggering the production of IL-17 and other effector molecules that promote inflammation and tumor cell proliferation(58). Intratumor microbiota can induce an immunosuppressive TME and promote lung cancer progression. Notably, reduced bacterial loads correlate with decreased Tregs, enhanced NK cell activity, and reduced lung cancer metastasis(59). This finding highlights the importance of microbiota-immune interactions in cancer progression. Studies have revealed a Treg cell-driven mechanism underlying lung-specific immunosuppression(60, 61). Additionally, Wang et al. reported that the tumor-resident fungus *Aspergillus sydowi* induces IL-1β secretion and MDSC activation via the β-glucan-mediated Dectin-1/CARD9 pathway, thereby advancing lung cancer progress(62). Low concentration of butyrate from the intratumor microbiome may promote lung cancer progression and metastasis by enhancing M2 macrophage polarization and function(63). In summary, the microbiome contributes to lung cancer through multiple biological pathways, including genotoxicity, inflammation, immune response, and angiogenesis.

3.2.2 Nasopharyngeal cancer

Nasopharyngeal carcinoma (NPC) originates from the nasopharyngeal mucosa as an epithelial carcinoma in NPC tumor tissues, microbiota is presented, with *Corynebacterium* and *Staphylococcus* being predominant (64, 65). The NPC microbiota plays a role in intratumoral infiltration and TME remodeling. Liu et al. reported a strong association between high bacterial load and reduced $CD8^+$ T cell infiltration, which contributes to an immunosuppressive environment in NPC(66). Furthermore, patients with a high bacterial load exhibited significant proliferation dependent on cell cycling. Oral-derived microbes were significantly enriched in the nasopharynx and closely associated with epithelial EBV infection(67). This finding suggests that blocking the translocation of microbes from the oral cavity to the

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nasopharynx could be a potential preventative intervention for NPC.

3.3 Reproductive system neoplasms

3.3.1 Cervical cancer

Intratumor microbiota may promote cervical carcinogenesis by inducing immune response drivers. Human papillomavirus (HPV) is established as the primary causative agent in cervical carcinogenesis. *Chlamydia trachomatis* is recognized as a cofactor of HPV in cervical cancer. Coinfections with *Chlamydia trachomatis* are more frequently observed in patients with invasive cervical cancers(68). Cervical cancer development involves a substantial decrease in *Lactobacillus* species and an increase in *Gardnerella vaginalis*, *Prevotella bivia*, *Porphyromonas spp.*, and *Streptococcus spp.*(69). *Lactobacillus* depletion may foster a proinflammatory environment, inducing malignant cell proliferation, and increasing HPV E6 and E7 oncogene expression, promoting cervical cancer development(70). *Lactobacillus iners*, a common species, appears to be an opportunistic pathogen(71). Colebert et al. illustrate that the intratumoral colonization of *Lactobacillus iners* in cervical cancer contributes to chemoradiotherapy resistance via L-lactate-mediated metabolic reprogramming, which is significantly associated with decreased patient survival rates. (72). Furthermore, higher levels of *Robiginitomaculum*, *Klebsiella*, *Micromonospora*, and *Microbispora* are linked to cervical cancer mortality, while *Methylobacter* levels show an inverse relationship(73). Microbiome relative abundance and tumor classification can predict cervical cancer prognosis.

3.3.2 Prostate cancer

Prostate cancer is the second most frequent malignancy among men worldwide(74). Current evidence indicates that the microbiota induces an inflammatory prostate microenvironment, promoting prostate cancer development and progression(75). Infection with *Propionibacterium* acnes activates the COX2-prostaglandin and plasminogen-matrix metalloproteinase pathways, triggering a strong inflammatory response(76). Additionally, Ma et al. found *Pediococcus pentosaceus*, *Listeria*

monocytogenes, and *Lactobacillus crispatus* in prostate cancer(77). Therefore, further investigation into microbiome interaction with prostate cancer cells is warranted.

3.4 Other cancers

3.4.1 Breast cancer

Breast cancer is the most common type of cancer in women. The metabolic status of breast cancer is highly plastic, and certain microbial genera are significantly correlated with metabolic activity in cancer. Researchers found that *Porphyromonas, Lacibacter, Ezakiella, Fusobacterium*, and *Pseudomonas* are more abundant in higher-stage breast tumors compared to lower-stage tumors and healthy breast microbiotas. These genera exhibit distinct features in normal and tumor tissues (78). Additionally, Robertson et al. discovered that breast cancer contains fungi, viruses, Chlamydiae, and parasites (Figure 2A)(79).

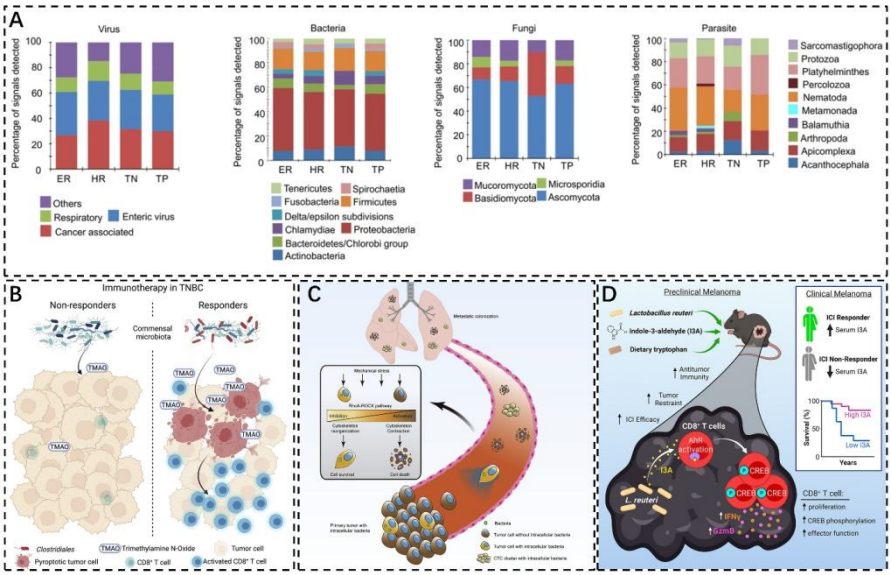

Figure 2. (A) Bar graphs showing breast cancer contain fungi, viruses, Chlamydiae, and parasites. Reproduced with permission(79). Copyright, 2021. (B) Schematic illustration of the microbial metabolite trimethylamine N-oxide promotes antitumor immunity in TNBC. Reproduced with permission(82). Copyright, 2022, Elsevier Inc. (C) Schematic illustration of tumor-resident intracellular microbiota promotes metastatic colonization in breast cancer. Reproduced with permission(78). Copyright, 2022, Elsevier Inc. (D) Schematic illustration of dietary tryptophan metabolite released

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by Lactobacillus reuteri facilitates ICI treatment. Reproduced with permission(10).Copyright, 2023, Elsevier Inc.

Intratumor microbiota may affect the tumor immune microenvironment. *Propionibacterium* is more abundant in healthy controls and NATs but is scarce in tumor tissues. Higher *Propionibacterium* levels correlate with increased T-cell activation and decreased oncogenic growth, suggesting that its absence could promote tumorigenesis by inhibiting an adaptive antitumor response and fostering a pro-tumorigenic environment. Besides directly affecting host immune responses, breast microbiota produce substances that can boost the antitumor immunity response. With a higher prevalence in non-breast cancer tissues, the *Streptococcus* genus generates cadaverine, which inhibits breast cancer invasion and epithelial-mesenchymal transition (80). Moreover, bile acids from the microbiota in breast tumors are associated with high cell proliferation and poorer survival rates(81). Shao et al. found that the microbial metabolite trimethylamine N-oxide activates the endoplasmic reticulum stress kinase PERK, inducing pyroptosis in malignant cells and strengthening the immune response against triple-negative breast cancer (TNBC) (Figure 2B)(82). Furthermore, in a mouse model of spontaneous breast tumors, the presence of bacteria induces a reorganization of the actin cytoskeleton in circulating tumor cells (CTC) in the blood. This restructuring enhances CTC's ability to withstand fluid shear stress, promoting CTC survival and breast cancer metastasis(Figure 2C)(78). Although present in low biomass, tumor-resident microbiota plays a crucial role in cancer metastasis, making their manipulation a potentially valuable avenue for advancing oncology care.

3.4.2 Melanoma

The cutaneous microbiota, with its potential role in melanoma development and treatment response, has emerged as a promising research area. Melanoma, a highly malignant skin tumor, has seen dramatically increased incidence rates globally (83). Compared to other tumors, such as bone, pancreatic, and breast, melanoma has a

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relatively lower bacterial biomass. The predominant bacterial genera in melanoma have been identified as *Acinetobacter*, *Actinomyces*, *Corynebacterium*, *Enterobacter*, *Roseomonas*, and *Streptococcus*(5).

Intratumor microbiota may inhibit T cell and NK cell activation, thereby prompting melanoma development. In a mouse melanoma model, antibiotic treatment in the lung reduced bacterial load, decreased regulatory T cells, and enhanced T cells and NK cell activation, resulting in fewer lung metastases. Furthermore, recent studies implied a link between *Corynebacterium* species and melanoma progression, possibly through an IL-17-dependent pathway. Zitvogel et al. reported a stage-associated increase in *Corynebacterium* among 27 patients with acral melanoma by analyzing culture-based skin swabs and the patients exhibited a higher frequency of Th17 cells. These findings highlight the potential impact of the skin microbiota on local cancers(84). Changes in microbiome composition affect immunotherapy treatment outcomes (85). The probiotic *Lactobacillus reuteri* , which colonizes and persists within melanoma, secretes dietary tryptophan catabolite indole-3-aldehyde, stimulating an immune response. Indole-3-aldehyde activates AhR signaling in $CD8^+$ T cells, promoting interferon-γ production and enhancing the efficacy of immune checkpoint inhibitors (Figure 2D)(10). Emerging evidence indicates that the composition and diversity of the skin microbiota critically modulate the efficacy of immune checkpoint inhibitors for skin cancer treatment.

4. Crosstalk between Intratumor Microbiota and TME

The relationship between the microbiota and tumors is highly complex, involving an intricate network of microbial infection, dysbiosis, tumorigenesis, and TME(86-88). The TME, encompassing tumor cells, immune cells, stromal cells, and a dense microvascular network, is the critical internal milieu essential for tumor cell existence and proliferation. Intratumor microbiota has been identified as a crucial component of TME(6). The unique features of TME including hypoxia, angiogenesis, low pH, and immunosuppression, facilitate microbial invasion and colonization. In turn, intratumor microbiota can affect the function of tumor and immune cells and their TME, playing

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a critical role in cancer initiation, development, and metastasis.

4.1 Effect of TME on Microbial Colonization

Several studies suggest that the microbiota migrates after cancer formation. Specific tumor microenvironments may facilitate microbial invasion and colonization. The hypoxic environment in tumors is highly favorable for the growth of facultative anaerobes and anammox bacteria, including *Clostridium perfringens*, *Escherichia coli*, *Listeria monocytogenes*, and *Bifidobacterium bifidu*m(89). Additionally, increased endothelial leakiness in tumor neovascularization facilitates microbiota entry into the tumor tissue through the bloodstream (90). Furthermore, constantly necrotic tumor tissue provides ample nutrients for microbial reproduction. Most cancer cells depend on aerobic glycolysis, known as the Warburg effect, to provide energy for tumor growth (91). Unlike most normal tissues, tumor cells tend to generate lactate from glucose even when oxygen is sufficient to support mitochondrial oxidative phosphorylation. The acidic environment promotes a highly immunosuppressive state in the TME. Deregulations in the tumor immune system may permit unhindered pathogen growth. These features create conducive conditions for microbiota colonization and survival.

4.2 Role of intratumor microbiota in TME

The International Agency for Cancer Research estimates that about 3.7×10^{30} microorganisms inhabit the earth, contributing to about 20% of human malignancies, with only 12 identified as human carcinogens(92). Microorganisms within tumors have a dual role. On the one hand, intratumor microbiota can promote tumor progression by inducing immunosuppression, inflammation, oncogenic signaling pathways, and host gene mutations. On the other hand, intratumor microbiota can enhance antitumor immunity and therapeutic efficacy by activating STING, promoting tertiary lymphoid structure maturation (93, 94), and presenting microbial antigens. The role of intratumor microbiota as a promoter or suppressor of tumors depends on its composition and abundance, tumor stage, and the host immune system response. Considering the diverse microbiota composition across tumors, targeting specific intratumor microbiota may

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enable precision diagnosis, treatment, and prognosis in clinical settings.

5. Potential Applications of Intratumor Microbiota in Cancer Therapy

Given the crucial role played by intratumor microbiota in cancer initiation and development, intratumor microbiota can be utilized to stimulate the immune response, thus enhancing the efficacy of immunotherapy. On the other hand, intratumor microbiota can be targeted to eliminate microorganisms at specific sites as targets, thereby enhancing precision cancer treatments.

5.1 Intratumor microbiota as immune enhancers in immunotherapy

In recent years, immunotherapy has become a critical component in cancer treatment(95-97). The role of intratumor microbiota in cancer immunotherapy is increasingly recognized as studies expand. As immune enhancers, intratumor microbiota can improve antitumor immune responses by remodeling TME, inducing pyroptosis, and prompting tumor and immune cells to present microbial antigens, thereby enhancing the efficacy of immune checkpoint inhibitors (ICI) (98). Intratumor microbiota is closely linked to the host immune system response, potentially resolving current dilemmas in immunotherapy.

Eliminating certain intratumor microbiota can convert a cold TME, thereby enhancing the immunotherapy efficacy. Qu et al. effectively reversed the cold TME by eliminating *Fusobacterium nucleatum*, which was then repurposed as an immune enhancer for triple-negative breast cancer (TNBC) immunotherapy. Furthermore, dead *Fusobacterium nucleatum* and non-cellular outer membrane vesicles served as immune enhancers, prompting dendritic cell maturation and T-cell infiltration, which significantly boosted the efficacy of TNBC immunotherapy (Figure 3A)(99). Gastric cancer associated with *Helicobacter pylori* infection exhibits a higher density of PD-L1+ cells and non-exhausted $CD8^+$ T cells, suggesting that *Helicobacter pylori* promotes a hot TME and is a favorable prognostic factor for gastric cancer immunotherapy (100). The accumulation of *Bifidobacterium bifidum* in tumors, which

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occurs in an interferon-dependent and STING pathway, facilitates anti-CD47 immunotherapy(101). Han et al. analyzed 1,296 intratumor microbiome genera across samples from the TCGA database, offering a comprehensive view of the links between the intratumor microbiome and immune features (Figure 3B)(102). Their findings revealed that *Eudoraea* increases the abundance of active immune cells in the TME, potentially improving immune checkpoint blockade (ICB) treatment outcomes.

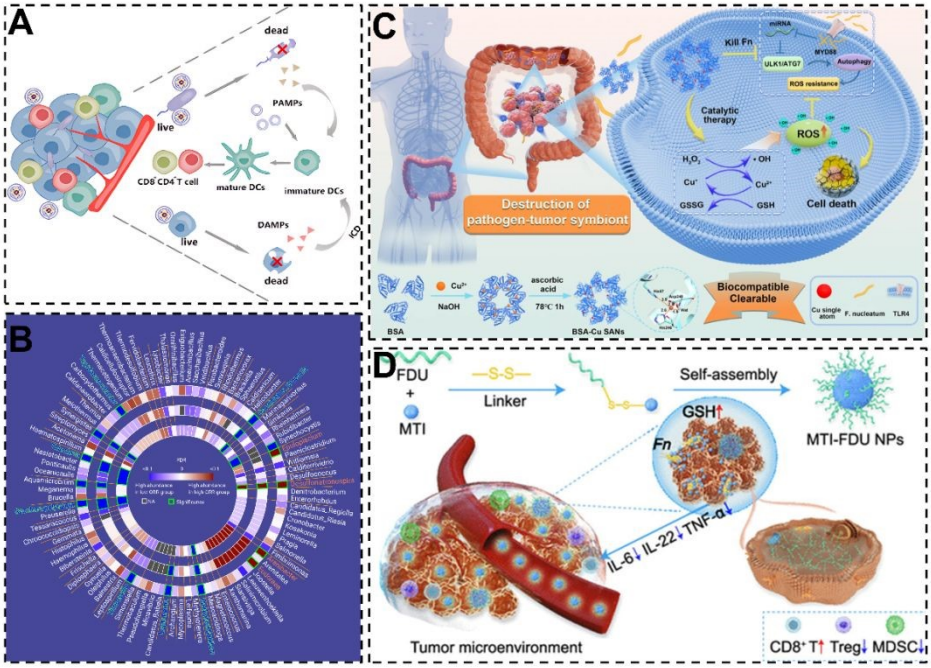

Figure 3. (A) Schematic depiction of intratumor bacteria reverse cold tumors for enhanced therapy of TNBC. Reproduced with permission (99). Copyright, 2023, American Chemical Society. (B) Associations between intratumor microbiome abundance and objective response rate at genus level, family level, and order level. Reproduced with permission (102). Copyright, 2023. (C) Schematic illustration of the synthesis of BSA-Cu SAN and its function of destroying pathogen-tumor symbionts for antitumor therapy. Reproduced with permission (126). Copyright 2023, Springer Protocols. (D) Schematic illustration of the synergistic target of intratumor microbiome and tumor via MTI-FDU. Reproduced with permission (135). Copyright 2023, American Chemical Society.

Intratumor microbiota may trigger pyroptosis, thereby enhancing the efficacy of

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immunotherapy. Pyroptosis, a regulated form of cell death, is mediated by the pore-forming, membrane-targeting gastrin family of proteins(103). Pyroptosis is a critical regulator in physiological processes such as inflammation, cell development, tissue homeostasis, and stress response(104). Shen et al. demonstrated that intravenous administration of *Listeria monocytogenes* induces gasdermin C-dependent pyroptosis, which modifies the immunosuppressive TME and enhances the efficacy of immunotherapy by prompting a robust antitumor immune response. (105). Sun et al. developed an oral bacterial pyroptosis amplifier for CRC. They utilized hyaluronic acid–coated *Shewanella oneidensis* MR-1 and engineered *Escherichia coli* for tumor-specific accumulation, leveraging hyaluronic acid's role in bacterial survival with the harsh gastrointestinal tract. This approach enhances oncolytic microbe-triggered pyroptosis, activates the gut mucosal immune response, and inhibits colon tumors and metastasis(106).

Furthermore, the display of intratumor bacteria peptide on host cell human leukocyte antigens (HLA) may enable the utilization of the intratumor microbiome in cancer immunotherapy. Microbial antigens, sharing antigenic epitopes with tumor antigens, recruit T cells specific to the microbe, which then recognize and eliminate tumor cells. Given bacterial antigens are considered non-autologous, they may serve as targets for immunotherapy to stimulate immune responses. Straussman et al. used 16S rRNA gene sequencing and HLA peptidomics to profile peptide libraries from intratumor bacteria, revealing that tumor cells can present peptides from these bacteria to provoke an immune response(107). Martin et al. also found that microbial peptides activate tumor-infiltrating lymphocytes(TILs) and peripheral blood memory cells, initiating an immune response(108). TILs are essential for the success of immunotherapy and improved survival in various tumors(109). Consequently, intratumor microbiota can trigger an immune response via microbial peptides.

5.2 Intratumor microbiota as new targets for drug delivery

Researchers are developing drug delivery systems to enhance the effectiveness of

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antitumor treatments(110-113). These systems, such as liposomes, hydrogels, and nanoparticles, offer biocompatibility, prolonged circulation, and high drug-loading capacity for anticancer drugs(114-116). Given the critical role of the intratumor microbiota in carcinogenesis, eliminating specific pathogenic microbiota (Table 1) and restoring tumor sensitivity to ICB therapy could enhance precision cancer treatments and prevent recurrence.

Table 1. The intratumor microbiota serves as target for nanodrug delivery.

	Nanoplatform	Microbiota	Work	Ref.
Delivery of antimicrobial agents	LipoAgTNZ	*Fusobacterium nucleatum, Escherichia coli Nissle.*	Eliminating intratumor bacteria in the primary tumor and liver metastases. Generating microbial neoantigens.	(121)
	MTZ/Ftn-DOX@HM	*Fusobacterium nucleatum*	Eliminating intratumor bacteria. Remodeling the ITME.	(122)
	BSA-Cu-SAN	*Fusobacterium nucleatum*	Disrupting the pathogen-tumor symbiosis	(126)
	N-CSs		Reversing Gem resistance. Generating •OH radicals.	(127)
	Au@BSA-CuPpIX	*Fusobacterium nucleatum*	Enhancing ROS-induced apoptosis and the therapeutic	(128)

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			efficacy of SDT for orthotopic CRC and inhibiting lung metastasis.	
	F. nucleatum-mimicking nanomedicine	*Fusobacterium nucleatum*	Killing intratumor bacteria Restoring ICB therapies.	(129)
Co-delivery of antitumor drugs and antimicrobial agents	sNP@G/IR	*Escherichia coli* Nissle 1917	Eliminating tumor-resident intracellular bacteria Augmenting drug delivery efficacy.	(134)
	MTI-FDU	*Fusobacterium nucleatum*	Achieving the dual target of the intratumor microbiota and tumor cells.	(135)
	PG-Pt-LA/CB (7)	*Fusobacterium nucleatum*	Eliminating *Fusobacterium nucleatum.* Enhancing chemotherapeutic efficacy.	(117)
	GC-DCPA-H2O		Eliminating intratumor microbiota. Enhancing chemotherapeutic efficacy.	(118)
	OLP/PP nanoassembly	*Fusobacterium nucleatum*	Eliminating *Fusobacterium nucleatum.*	(119)

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	Inhibiting tumor growth.

5.2.1 Delivery of antimicrobial agents

Antibiotics are routinely used in clinical settings for bacterial infections. However, a single antibiotic's ability to permeate tumor cells and address bacterial infections is impeded by cell membrane barriers and the complex TME. Systemic administration of antibiotics targeting pathogenic bacteria in tumor may disrupt other microbiota in the body. Hence, precise antibiotic delivery to deplete intratumor pathogenic microbiota without upsetting systemic microbiota imbalance is essential. Drug carrier nanoparticles can selectively target microbiota colonized within tumor cells(120). Huang et al. recently developed liposome-encapsulated silver-tinidazole complex to eliminate intratumor bacteria using a remote loading technique. The treatment generates microbial neoantigens that enhance immune system recognition of both infected and uninfected tumor cells, triggering an antitumor immune response(121). Yang et al. also developed a biomimetic nanovehicle mimicking *Fusobacterium nucleatum* for targeted antibiotic delivery(122). This approach effectively eliminates intratumor *Fusobacterium nucleatum* and restores tumor sensitivity to ICB therapy.

The misuse of antibiotics has fostered the emergence of multidrug-resistant strains, complicating antibiotic-dependent treatments(123). Recently, nanozymes have emerged as alternatives to traditional antibiotics, catalyzing reactive oxygen species (ROS) generation to kill bacteria or disrupt biofilms without inducing drug resistance(124, 125). Qin et al. designed protein-supported copper single-atom nanozymes (BSA-Cu-SAN) that can passively target tumors, producing ROS and depleting glutathione to promote cancer cell apoptosis. This strategy efficiently eliminates intratumor *Fusobacterium nucleatum* and disrupts pathogen-tumor symbionts, blocking the interaction between intratumor microbiota and tumors (Figure 3C)(126). Gao et al. used nitrogen-doped carbon nanospheres (N-CSs) as nanozymes to generate hydroxyl radicals for catalytic tumor therapy. By inhibiting cytidine

deaminase, the N-CSs effectively reverse Gem resistance induced by bacterial cytidine deaminase in mouse models(127). Furthermore, Qin et al. designed an albumin-based nanoplatform responsive to ultrasonic stimulation. This strategy eradicated *Fusobacterium nucleatum* and promoted cancer cell apoptosis by increasing ROS levels(128).

In addition, nanomedicines that mimic bacteria have been used to target intratumor bacteria specifically, preserving the intestinal microbiota. Chen et al. created a nanomedicine mimicking *Fusobacterium nucleatum* combining its cytoplasmic membranes with liposomes containing polymyxins. This treatment has shown promise in restoring sensitivity to ICB therapy in tumors colonized by *Fusobacterium nucleatum*(129).

5.2.2 Co-delivery of antitumor drug and antimicrobial agents

Some chemotherapy drugs, known to induce immunogenic cell death (ICD), can enhance the efficacy of tumor immunotherapy(130). Observations indicate that microbiota may contribute to resistance to chemotherapeutic drugs (131-133). Specifically, *Gammaproteobacteria* residing in tumors can convert the chemotherapeutic drug Gem into an inactive form, reducing chemotherapy efficacy. Combining chemotherapy with immunotherapy to eliminate intratumor microbiota may enhance the efficacy of tumor treatment. Nano-drug delivery systems hold promise for targeting lesion sites and achieving intelligent, sustained drug release. Therefore, integrating antimicrobial agents and chemotherapeutic drugs into nano-drug delivery systems could target the elimination of intratumor microbiota and overcome drug resistance, enhancing the efficacy of antitumor therapy.

Eliminating intratumor microbiota can increase drug delivery efficacy and, subsequently, the release of antitumor drugs, achieving a synergistic antitumor effect by dual targeting of intratumor microbiota and tumors. Wang et al. designed a dual-cascade responsive nanoparticle (sNP@G/IR) containing Gem and a photothermal agent (IR1048) to enhance antitumor efficacy, with the polymer core serving an

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antimicrobial function. In addition, the hyperthermic effect of IR1048 aids in further eliminating tumors and bacteria(134). Ma et al. designed metronidazole–fluorouridine nanoparticles (MTI-FDU) to achieve a synergistic antitumor effect. The nanoparticles metronidazole target intratumor bacteria with minimal disruption to gut microbial homeostasis (Figure 3D)(135). Li et al. developed size-tunable nanogels that integrate Zinc-imidazolate frameworks with encapsulated doxorubicin (DOX) and folate grafting (f-ZIFD), combined with metronidazole. The sequential release of f-ZIFD nanoparticles from NGs promotes effective tumor penetration and precise tumor cell targeting, while the acidic-triggered intracellular release of doxorubicin enhances the antitumor effect(136).

5.3 Intratumor microbiota as an anticancer therapeutic agent

In the past decades, engineered strains such as *Salmonella* and *Clostridium* have effectively slowed tumor growth and metastasis(137), enhancing survival in preclinical models and clinical cases. Bacterial-based therapies, often used as drug carriers, benefit from nanotechnology(138), synthetic bioengineering(139), and genetic engineering (140) to attenuate bacteria and enhance drug efficacy in oncology treatments. Recently, intratumor bacteria have emerged as promising natural anticancer therapeutic agents. Miyako et al. isolated three types of intratumor bacteria-*Rhodopseudomonas palustris*, *Proteus mirabilis,* and a complex bacterium of these two from CRC tissue. These isolated bacteria possess inherent biocompatibility and potent immunogenic anticancer efficacies. They selectively grow and proliferate within the tumor environment, effectively prompting immune cells to infiltrate and eliciting robust anticancer responses in mice(141). *Staphylococcus epidermidis,* producing 6-N-hydroxyaminopurine, inhibits skin tumor growth(142). Modifying these bacteria to enhance their tumor-targeting capabilities represents a promising new research direction.

6. Conclusions

Recent studies highlight the significant role of intratumor microbiota in influencing

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tumor therapy outcomes. The microbiota residing within tumors can alter the tumor microenvironment, potentially impacting therapeutic efficacy. Here, we summarize the characteristics and interactions of intratumor microbiota with various cancers, emphasizing their composition and the critical roles they play. Intratumor microbiota consists of diverse microbial communities, which can differ markedly between tumor types and even between patients with the same cancer. For example, certain bacterial species may be more prevalent in specific tumors, influencing tumor behavior and patient response to treatment. Research has shown that these microbes can modulate immune responses, affect drug metabolism, and even alter the tumor microenvironment, potentially enhancing or inhibiting therapeutic effects.

The therapeutic applications of intratumor microbiota are emerging as a promising area of research. Potential strategies include: 1. Microbiota-based therapies: Modulating the intratumor microbiota through probiotics or targeted therapies to enhance immune responses. 2. Biomarker development: Identifying specific microbial signatures that correlate with treatment responses could help tailor therapies to individual patients. 3. Combination therapies: Integrating microbiota modulation with existing cancer therapies, such as immunotherapy or chemotherapy, may enhance treatment efficacy. However, it is crucial to maximize the benefits of intratumor microbiota while minimizing potential adverse effects. This requires a thorough understanding of how these microbial communities interact with cancer therapies.

Several limitations hinder the advancement of research on intratumor microbiota. First, the low abundance of intratumor microbiota presents challenges in obtaining adequate samples from tumor tissues. This scarcity can lead to difficulties in accurately assessing microbial diversity and composition. Secondly, current sequencing methods, such as 16S rRNA gene sequencing, primarily provide relative abundance data. This can lead to misunderstandings regarding microbial community structures and interactions. Advanced techniques like metagenomic sequencing are needed for more comprehensive insights. Additionally， enhancing the detection rate of intratumoral

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bacteria presents a significant challenge, as the inability to accurately identify bacterial components undermines the comprehensive assessment of microbial diversity and composition, while exacerbating contamination artifacts. To address this issue, the use of microbial DNA pre-amplification through multiple displacement amplification (MDA) lowers the detection threshold to an abundance of 0.1%. Simultaneously, the application of MolYsis™ reagent kits facilitates the selective lysis of eukaryotic cells while preserving intact microbial cells. Complementary techniques, such as laser microdissection (LMD), enable precise isolation of tumor parenchymal regions, thereby reducing stromal contamination and ensuring data fidelity in studies of microbe-host interactions. Finally, establishing suitable cell and small animal models for studying intratumor microbiota is essential for basic and preclinical research. The advancement of patient-derived organoid-bacteria coculture systems offers a promising approach to address this requirement. The inherent heterogeneity of tumors and their associated microbiota complicates model development, requiring innovative approaches to replicate tumor conditions accurately.

Despite existing preclinical evidence linking intratumor microbial modulation with immunotherapy outcomes, further research is necessary to validate these findings in clinical settings. To enhance our understanding, future investigations should focus on exploring the specific mechanisms through which intratumor microbiota influence tumor progression, metastasis, and therapeutic responses. And, utilizing multi-omics technologies and refined reference databases to ensure comprehensive and accurate analyses of intratumor microbiota. By integrating insights from microbiota research with conventional cancer therapies, we may pave the way for more effective and personalized cancer treatment strategies.

DECLARATION OF INTERESTS

All authors confirmed no declarations of interest.

AUTHOR CONTRIBUTIONS

Huiling Liu: Writing – original draft, Writing – review & editing. **Zhonghui Luo:** Writing – original draft, Writing – review & editing. **Fangzhen Luo**: Writing – original draft, Writing – review & editing. **Xilian Wang**: Writing – original draft, Writing – review & editing. **Hua Wei**: Writing – original draft, Writing – review & editing. **Cui-Yun Yu**: Writing – original draft, Writing – review & editing.

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DECLARATION OF GENERATIVE AI AND AI-ASSISTED TECHNOLOGIES IN THE WRITING PROCESS

During the preparation of this work, the authors used ChatGPT in order to improve the grammatical structure in some paragraphs; all the research cited came from PubMed. The authors reviewed all the references cited to ensure the accurate representation of the original research findings. After using ChatGPT, the authors reviewed and edited the content as needed and take full responsibility for the content of the publication.

References

1. Crosby D, Bhatia S, Brindle KM, Coussens LM, Dive C, Emberton M, et al. Early detection of cancer. Science. 2022;375(6586):1244-+.

2. Borcherding N, Brestoff JR. The power and potential of mitochondria transfer. Nature. 2023;623(7986):283-91.

3. Wen L, Mu W, Lu H, Wang X, Fang J, Jia Y, et al. Porphyromonas gingivalis Promotes

View Article Online
DOI: 10.1039/D5PM00045A

Oral Squamous Cell Carcinoma Progression in an Immune Microenvironment. Journal of Dental Research. 2020;99(6):666-75.

4. Taglialegna A. When Helicobacter pylori spells gastric cancer. Nature Reviews Microbiology. 2023;21(10):628-.

5. Nejman D, Livyatan I, Fuks G, Gavert N, Zwang Y, Geller LT, et al. The human tumor microbiome is composed of tumor type-specific intracellular bacteria. Science. 2020;368(6494):973-+.

6. Galeano Niño JL, Wu H, LaCourse KD, Kempchinsky AG, Baryiames A, Barber B, et al. Effect of the intratumoral microbiota on spatial and cellular heterogeneity in cancer. Nature. 2022;611(7937):810-7.

7. Narunsky-Haziza L, Sepich-Poore GD, Livyatan I, Asraf O, Martino C, Nejman D, et al. Pan-cancer analyses reveal cancer-type-specific fungal ecologies and bacteriome interactions. Cell. 2022;185(20):3789-806.e17.

8. Ma Y, Chen H, Li H, Zheng M, Zuo X, Wang W, et al. Intratumor microbiome-derived butyrate promotes lung cancer metastasis. Cell Reports Medicine. 2024;5(4).

9. Battaglia TW, Mimpen IL, Traets JJH, van Hoeck A, Zeverijn LJ, Geurts BS, et al. A pan-cancer analysis of the microbiome in metastatic cancer. Cell. 2024;187(9):2324-35.e19.

10. Bender MJ, McPherson AC, Phelps CM, Pandey SP, Laughlin CR, Shapira JH, et al. Dietary tryptophan metabolite released by intratumoral Lactobacillus reuteri facilitates immune checkpoint inhibitor treatment. Cell. 2023;186(9):1846-62.e26.

11. Thrift AP. Global burden and epidemiology of Barrett oesophagus and oesophageal

View Article Online
DOI: 10.1039/D5PM00045A

cancer. Nature Reviews Gastroenterology & Hepatology. 2021;18(6):432-43.

12. Greathouse KL, Stone JK, Vargas AJ, Choudhury A, Padgett RN, White JR, et al. Co-enrichment of cancer-associated bacterial taxa is correlated with immune cell infiltrates in esophageal tumor tissue. Scientific Reports. 2024;14(1):2574.

13. Wu H, Leng XF, Liu QS, Mao TQ, Jiang T, Liu YQ, et al. Intratumoral Microbiota Composition Regulates Chemoimmunotherapy Response in Esophageal Squamous Cell Carcinoma. Cancer Res. 2023;83(18):3131-44.

14. Liang M, Liu Y, Zhang Z, Yang H, Dai N, Zhang N, et al. Fusobacterium nucleatum induces MDSCs enrichment via activation the NLRP3 inflammosome in ESCC cells, leading to cisplatin resistance. Annals of Medicine. 2022;54(1):989-1003.

15. Nomoto D, Baba Y, Liu Y, Tsutsuki H, Okadome K, Harada K, et al. Fusobacterium nucleatum promotes esophageal squamous cell carcinoma progression via the NOD1/RIPK2/NF-κB pathway. Cancer Letters. 2022;530:59-67.

16. Guo S, Chen F, Li L, Dou S, Li Q, Huang Y, et al. Intracellular Fusobacterium nucleatum infection increases METTL3-mediated m6A methylation to promote the metastasis of esophageal squamous cell carcinoma. Journal of Advanced Research. 2023.

17. Zhang J-W, Zhang D, Yin H-S, Zhang H, Hong K-Q, Yuan J-P, et al. Fusobacterium nucleatum

promotes esophageal squamous cell carcinoma progression and chemoresistance by enhancing the secretion of chemotherapy-induced senescence-associated secretory phenotype via activation of DNA damage response pathway. Gut Microbes. 2023;15(1).

View Article Online
DOI: 10.1039/D5PM00045A

18. Li Y, Xing S, Chen F, Li Q, Dou S, Huang Y, et al. Intracellular Fusobacterium nucleatum infection attenuates antitumor immunity in esophageal squamous cell carcinoma. Nature Communications. 2023;14(1):5788.

19. Navashenaq JG, Shabgah AG, Banach M, Jamialahmadi T, Penson PE, Johnston TP, et al. The interaction of Helicobacter pylori with cancer immunomodulatory stromal cells: New insight into gastric cancer pathogenesis. Seminars in Cancer Biology. 2022;86:951-9.

20. Fu K, Cheung AHK, Wong CC, Liu W, Zhou Y, Wang F, et al. Streptococcus anginosus promotes gastric inflammation, atrophy, and tumorigenesis in mice. Cell. 2024;187(4):882-96.e17.

21. Yuan L, Pan L, Wang Y, Zhao J, Fang L, Zhou Y, et al. Characterization of the landscape of the intratumoral microbiota reveals that Streptococcus anginosus increases the risk of gastric cancer initiation and progression. Cell Discov. 2024;10(1):117.

22. Murata-Kamiya N, Hatakeyama M. Helicobacter pylori-induced DNA double-stranded break in the development of gastric cancer. Cancer Science. 2022;113(6):1909-18.

23. Yong X, Tang B, Xiao Y-F, Xie R, Qin Y, Luo G, et al. Helicobacter pylori upregulates Nanog and Oct4 via Wnt/β-catenin signaling pathway to promote cancer stem cell-like properties in human gastric cancer. Cancer Letters. 2016;374(2):292-303.

24. Lim MCC, Jantaree P, Naumann M. The conundrum of Helicobacter pylori-associated apoptosis in gastric cancer. Trends in Cancer. 2023;9(8):679-90.

25. Imai S, Ooki T, Murata-Kamiya N, Komura D, Tahmina K, Wu W, et al. Helicobacter pylori CagA elicits BRCAness to induce genome instability that may underlie bacterial gastric

View Article Online
DOI: 10.1039/D5PM00045A

carcinogenesis. Cell Host & Microbe. 2021;29(6):941-58.e10.

26. Salvatori S, Marafini I, Laudisi F, Monteleone G, Stolfi C. Helicobacter pylori and Gastric Cancer: Pathogenetic Mechanisms. International Journal of Molecular Sciences. 2023;24(3):2895.

27. Park EM, Chelvanambi M, Bhutiani N, Kroemer G, Zitvogel L, Wargo JA. Targeting the gut and tumor microbiota in cancer. Nature Medicine. 2022;28(4):690-703.

28. Arima K, Zhong R, Ugai T, Zhao M, Haruki K, Akimoto N, et al. Western-Style Diet, pks Island-Carrying Escherichia coli, and Colorectal Cancer: Analyses From Two Large Prospective Cohort Studies. Gastroenterology. 2022;163(4):862-74.

29. Pleguezuelos-Manzano C, Puschhof J, Rosendahl Huber A, van Hoeck A, Wood HM, Nomburg J, et al. Mutational signature in colorectal cancer caused by genotoxic pks+ E. coli. Nature. 2020;580(7802):269-73.

30. Chen B, Ramazzotti D, Heide T, Spiteri I, Fernandez-Mateos J, James C, et al. Contribution of pks+ E. coli mutations to colorectal carcinogenesis. Nature Communications. 2023;14(1).

31. Wilson MR, Jiang Y, Villalta PW, Stornetta A, Boudreau PD, Carrá A, et al. The human gut bacterial genotoxin colibactin alkylates DNA. Science. 2019;363(6428).

32. He Z, Gharaibeh RZ, Newsome RC, Pope JL, Dougherty MW, Tomkovich S, et al. Campylobacter jejuni promotes colorectal tumorigenesis through the action of cytolethal distending toxin. Gut. 2019;68(2):289-300.

33. Lopez LR, Bleich RM, Arthur JC. Microbiota Effects on Carcinogenesis: Initiation,

View Article Online
DOI: 10.1039/D5PM00045A

Promotion, and Progression. Annual Review of Medicine. 2021;72(1):243-61.

34. Zepeda-Rivera M, Minot SS, Bouzek H, Wu H, Blanco-Míguez A, Manghi P, et al. A distinct Fusobacterium nucleatum clade dominates the colorectal cancer niche. Nature. 2024.

35. Kong C, Yan X, Zhu Y, Zhu H, Luo Y, Liu P, et al. Fusobacterium Nucleatum Promotes the Development of Colorectal Cancer by Activating a Cytochrome P450/Epoxyoctadecenoic Acid Axis via TLR4/Keap1/NRF2 Signaling. Cancer Res. 2021;81(17):4485-98.

36. Cui W, Guo M, Liu D, Xiao P, Yang C, Huang H, et al. Gut microbial metabolite facilitates colorectal cancer development via ferroptosis inhibition. Nature Cell Biology. 2024;26(1):124-37.

37. Feng M, Pan Y, Kong R, Shu S. Therapy of Primary Liver Cancer. The Innovation. 2020;1(2).

38. Hepatocellular carcinoma. Nature Reviews Disease Primers. 2021;7(1):7.

39. Komiyama S, Yamada T, Takemura N, Kokudo N, Hase K, Kawamura YI. Profiling of tumour-associated microbiota in human hepatocellular carcinoma. Scientific Reports. 2021;11(1):10589.

40. Chai X, Wang J, Li H, Gao C, Li S, Wei C, et al. Intratumor microbiome features reveal antitumor potentials of intrahepatic cholangiocarcinoma. Gut Microbes. 2022;15(1).

41. <transjugular intrahepatic portosyste source jama so 2017 feb 28 317 8 880.pdf>.

42. Liu B, Zhou Z, Jin Y, Lu J, Feng D, Peng R, et al. Hepatic stellate cell activation and senescence induced by intrahepatic microbiota disturbances drive progression of liver cirrhosis toward hepatocellular carcinoma. Journal for ImmunoTherapy of Cancer. 2022;10(1).

View Article Online
DOI: 10.1039/D5PM00045A

43. Sun L, Ke X, Guan A, Jin B, Qu J, Wang Y, et al. Intratumoural microbiome can predict the prognosis of hepatocellular carcinoma after surgery. Clinical and Translational Medicine. 2023;13(7):e1331.

44. Xue C, Jia J, Gu X, Zhou L, Lu J, Zheng Q, et al. Intratumoral bacteria interact with metabolites and genetic alterations in hepatocellular carcinoma. Signal Transduction and Targeted Therapy. 2022;7(1).

45. Geller LT, Barzily-Rokni M, Danino T, Jonas OH, Shental N, Nejman D, et al. Potential role of intratumor bacteria in mediating tumor resistance to the chemotherapeutic drug gemcitabine. Science. 2017;357(6356):1156-60.

46. Pushalkar S, Hundeyin M, Daley D, Zambirinis CP, Kurz E, Mishra A, et al. The Pancreatic Cancer Microbiome Promotes Oncogenesis by Induction of Innate and Adaptive Immune Suppression. Cancer Discovery. 2018;8(4):403-16.

47. Hezaveh K, Shinde RS, Klötgen A, Halaby MJ, Lamorte S, Ciudad MT, et al. Tryptophan-derived microbial metabolites activate the aryl hydrocarbon receptor in tumor-associated macrophages to suppress anti-tumor immunity. Immunity. 2022;55(2):324-40.e8.

48. Yang X, Zhang Z, Shen X, Xu J, Weng Y, Wang W, et al. Clostridium butyricum and its metabolite butyrate promote ferroptosis susceptibility in pancreatic ductal adenocarcinoma. Cell Oncol (Dordr). 2023;46(6):1645-58.

49. Stasiewicz M, Karpiński TM. The oral microbiota and its role in carcinogenesis. Seminars in Cancer Biology. 2022;86:633-42.

50. Tan Q, Ma X, Yang B, Liu Y, Xie Y, Wang X, et al. Periodontitis pathogen

View Article Online
DOI: 10.1039/D5PM00045A

Porphyromonas gingivalis

promotes pancreatic tumorigenesis via neutrophil elastase from tumor-associated neutrophils. Gut Microbes. 2022;14(1).

51. Saba E, Farhat M, Daoud A, Khashan A, Forkush E, Menahem NH, et al. Oral bacteria accelerate pancreatic cancer development in mice. Gut. 2024.

52. Alam A, Levanduski E, Denz P, Villavicencio HS, Bhatta M, Alhorebi L, et al. Fungal mycobiome drives IL-33 secretion and type 2 immunity in pancreatic cancer. Cancer Cell. 2022;40(2):153-67.e11.

53. Aykut B, Pushalkar S, Chen R, Li Q, Abengozar R, Kim JI, et al. The fungal mycobiome promotes pancreatic oncogenesis via activation of MBL. Nature. 2019;574(7777):264-7.

54. Riquelme E, Zhang Y, Zhang L, Montiel M, Zoltan M, Dong W, et al. Tumor Microbiome Diversity and Composition Influence Pancreatic Cancer Outcomes. Cell. 2019;178(4):795-806.e12.

55. Ghaddar B, Biswas A, Harris C, Omary MB, Carpizo DR, Blaser MJ, et al. Tumor microbiome links cellular programs and immunity in pancreatic cancer. Cancer Cell. 2022;40(10):1240-53.e5.

56. Thai AA, Solomon BJ, Sequist LV, Gainor JF, Heist RS. Lung cancer. The Lancet. 2021;398(10299):535-54.

57. Goto T. Microbiota and lung cancer. Seminars in Cancer Biology. 2022;86:1-10.

58. Jin C, Lagoudas GK, Zhao C, Bullman S, Bhutkar A, Hu B, et al. Commensal

View Article Online
DOI: 10.1039/D5PM00045A

Microbiota Promote Lung Cancer Development via γδ T Cells. Cell. 2019;176(5):998-1013.e16.

59. Le Noci V, Guglielmetti S, Arioli S, Camisaschi C, Bianchi F, Sommariva M, et al. Modulation of Pulmonary Microbiota by Antibiotic or Probiotic Aerosol Therapy: A Strategy to Promote Immunosurveillance against Lung Metastases. Cell Reports. 2018;24(13):3528-38.

60. Zagorulya M, Yim L, Morgan DM, Edwards A, Torres-Mejia E, Momin N, et al. Tissue-specific abundance of interferon-gamma drives regulatory T cells to restrain DC1-mediated priming of cytotoxic T cells against lung cancer. Immunity. 2023;56(2):386-405.e10.

61. Watson MJ, Vignali PDA, Mullett SJ, Overacre-Delgoffe AE, Peralta RM, Grebinoski S, et al. Metabolic support of tumour-infiltrating regulatory T cells by lactic acid. Nature. 2021;591(7851):645-51.

62. Liu N-N, Yi C-X, Wei L-Q, Zhou J-A, Jiang T, Hu C-C, et al. The intratumor mycobiome promotes lung cancer progression via myeloid-derived suppressor cells. Cancer Cell. 2023;41(11):1927-44.e9.

63. Ma Y, Chen H, Li H, Zheng M, Zuo X, Wang W, et al. Intratumor microbiome-derived butyrate promotes lung cancer metastasis. Cell Reports Medicine. 2024.

64. Jin S, Li R, Chen M-Y, Yu C, Tang L-Q, Liu Y-M, et al. Single-cell transcriptomic analysis defines the interplay between tumor cells, viral infection, and the microenvironment in nasopharyngeal carcinoma. Cell Research. 2020;30(11):950-65.

65. He J, Liu L, Tang F, Zhou Y, Liu H, Lu C, et al. Paradoxical effects of DNA tumor virus oncogenes on epithelium-derived tumor cell fate during tumor progression and chemotherapy response. Signal Transduction and Targeted Therapy. 2021;6(1):408.

View Article Online
DOI: 10.1039/D5PM00045A

66. Qiao H, Tan X-R, Li H, Li J-Y, Chen X-Z, Li Y-Q, et al. Association of Intratumoral Microbiota With Prognosis in Patients With Nasopharyngeal Carcinoma From 2 Hospitals in China. JAMA Oncology. 2022;8(9).

67. Liao Y, Wu Y-X, Tang M, Chen Y-W, Xie J-R, Du Y, et al. Microbes translocation from oral cavity to nasopharyngeal carcinoma in patients. Nature Communications. 2024;15(1).

68. Koster S, Gurumurthy RK, Kumar N, Prakash PG, Dhanraj J, Bayer S, et al. Modelling Chlamydia and HPV co-infection in patient-derived ectocervix organoids reveals distinct cellular reprogramming. Nature Communications. 2022;13(1):1030.

69. Nikitina VP, Zykova TA, Shevyakova EA, Zhenilo OE, Verenikina EV, Ivanova VA, et al. Vaginal biocenosis in patients with gynecological cancers. Journal of Clinical Oncology. 2021;39(15_suppl):e17574-e.

70. Kyrgiou M, Moscicki A-B. Vaginal microbiome and cervical cancer. Seminars in Cancer Biology. 2022;86:189-98.

71. Zheng N, Guo R, Wang J, Zhou W, Ling Z. Contribution of Lactobacillus iners to Vaginal Health and Diseases: A Systematic Review. Frontiers in Cellular and Infection Microbiology. 2021;11.

72. Colbert LE, El Alam MB, Wang R, Karpinets T, Lo D, Lynn EJ, et al. Tumor-resident Lactobacillus iners confer chemoradiation resistance through lactate-induced metabolic rewiring. Cancer Cell. 2023;41(11):1945-62.e11.

73. Jiang L, Duan B, Jia P, Zhang Y, Yan X. The Role of Intratumor Microbiomes in Cervical Cancer Metastasis. Cancers. 2023;15(2).

View Article Online
DOI: 10.1039/D5PM00045A

74. Rastogi I, Muralidhar A, McNeel DG. Vaccines as treatments for prostate cancer. Nature Reviews Urology. 2023;20(9):544-59.

75. Sfanos KS, Yegnasubramanian S, Nelson WG, De Marzo AM. The inflammatory microenvironment and microbiome in prostate cancer development. Nature Reviews Urology. 2018;15(1):11-24.

76. Radej S, Szewc M, Maciejewski R. Prostate Infiltration by Treg and Th17 Cells as an Immune Response to Propionibacterium acnes Infection in the Course of Benign Prostatic Hyperplasia and Prostate Cancer. International Journal of Molecular Sciences. 2022;23(16):8849.

77. Ma J, Gnanasekar A, Lee A, Li WT, Haas M, Wang-Rodriguez J, et al. Influence of Intratumor Microbiome on Clinical Outcome and Immune Processes in Prostate Cancer. Cancers. 2020;12(9).

78. Fu A, Yao B, Dong T, Chen Y, Yao J, Liu Y, et al. Tumor-resident intracellular microbiota promotes metastatic colonization in breast cancer. Cell. 2022;185(8):1356-72.e26.

79. Banerjee S, Wei Z, Tian T, Bose D, Shih NNC, Feldman MD, et al. Prognostic correlations with the microbiome of breast cancer subtypes. Cell Death & Disease. 2021;12(9):831.

80. Kovács T, Mikó E, Vida A, Sebő É, Toth J, Csonka T, et al. Cadaverine, a metabolite of the microbiome, reduces breast cancer aggressiveness through trace amino acid receptors. Scientific Reports. 2019;9(1):1300.

81. Tang W, Putluri V, Ambati CR, Dorsey TH, Putluri N, Ambs S. Liver- and Microbiome-

View Article Online
DOI: 10.1039/D5PM00045A

derived Bile Acids Accumulate in Human Breast Tumors and Inhibit Growth and Improve Patient Survival. Clinical Cancer Research. 2019;25(19):5972-83.

82. Wang H, Rong X, Zhao G, Zhou Y, Xiao Y, Ma D, et al. The microbial metabolite trimethylamine N-oxide promotes antitumor immunity in triple-negative breast cancer. Cell Metabolism. 2022;34(4):581-94.e8.

83. Long GV, Swetter SM, Menzies AM, Gershenwald JE, Scolyer RA. Cutaneous melanoma. The Lancet. 2023;402(10400):485-502.

84. Routy B, Jackson T, Mählmann L, Baumgartner CK, Blaser M, Byrd A, et al. Melanoma and microbiota: Current understanding and future directions. Cancer Cell. 2024;42(1):16-34.

85. Björk JR, Bolte LA, Maltez Thomas A, Lee KA, Rossi N, Wind TT, et al. Longitudinal gut microbiome changes in immune checkpoint blockade-treated advanced melanoma. Nature Medicine. 2024;30(3):785-96.

86. Wong-Rolle A, Wei HK, Zhao C, Jin C. Unexpected guests in the tumor microenvironment: microbiome in cancer. Protein & Cell. 2020;12(5):426-35.

87. York A. Tumour-specific microbiomes. Nature Reviews Microbiology. 2020;18(8):413-.

88. Silveira MAD, Bilodeau S, Greten TF, Wang XW, Trinchieri G. The gut–liver axis: host microbiota interactions shape hepatocarcinogenesis. Trends in Cancer. 2022;8(7):583-97.

89. Zhang X, Yu D, Wu D, Gao X, Shao F, Zhao M, et al. Tissue-resident Lachnospiraceae family bacteria protect against colorectal carcinogenesis by promoting tumor immune surveillance. Cell Host & Microbe. 2023;31(3):418-32.e8.

View Article Online
DOI: 10.1039/D5PM00045A

90. Wei X, Chen Y, Jiang X, Peng M, Liu Y, Mo Y, et al. Mechanisms of vasculogenic mimicry in hypoxic tumor microenvironments. Molecular Cancer. 2021;20(1).

91. Pouysségur J, Marchiq I, Parks SK, Durivault J, Ždralević M, Vucetic M. 'Warburg effect' controls tumor growth, bacterial, viral infections and immunity – Genetic deconstruction and therapeutic perspectives. Seminars in Cancer Biology. 2022;86:334-46.

92. Zhou P, Hu Y, Wang X, Shen L, Liao X, Zhu Y, et al. Microbiome in cancer: An exploration of carcinogenesis, immune responses and immunotherapy. Frontiers in Immunology. 2022;13.

93. Overacre-Delgoffe AE, Bumgarner HJ, Cillo AR, Burr AHP, Tometich JT, Bhattacharjee A, et al. Microbiota-specific T follicular helper cells drive tertiary lymphoid structures and anti-tumor immunity against colorectal cancer. Immunity. 2021;54(12):2812-24.e4.

94. Cabrita R, Lauss M, Sanna A, Donia M, Skaarup Larsen M, Mitra S, et al. Tertiary lymphoid structures improve immunotherapy and survival in melanoma. Nature. 2020;577(7791):561-5.

95. Cao LL, Kagan JC. Targeting innate immune pathways for cancer immunotherapy. Immunity. 2023;56(10):2206-17.

96. Yi M, Li T, Niu M, Mei Q, Zhao B, Chu Q, et al. Exploiting innate immunity for cancer immunotherapy. Molecular Cancer. 2023;22(1).

97. Lee D, Huntoon K, Wang Y, Jiang W, Kim BYS. Harnessing Innate Immunity Using Biomaterials for Cancer Immunotherapy. Advanced Materials. 2021;33(27):2007576.

View Article Online
DOI: 10.1039/D5PM00045A

98. Gao Y, Bi D, Xie R, Li M, Guo J, Liu H, et al. Correction To: Fusobacterium nucleatum enhances the efficacy of PD-L1 blockade in colorectal cancer. Signal Transduction and Targeted Therapy. 2021;6(1):434.

99. 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. Journal of the American Chemical Society. 2023;145(48):26296-307.

100. Jia K, Chen Y, Xie Y, Wang X, Hu Y, Sun Y, et al. Helicobacter pylori and immunotherapy for gastrointestinal cancer. The Innovation. 2024;5(2).

101. Shi Y, Zheng W, Yang K, Harris KG, Ni K, Xue L, et al. Intratumoral accumulation of gut microbiota facilitates CD47-based immunotherapy via STING signaling. Journal of Experimental Medicine. 2020;217(5).

102. Zhang Z, Gao Q, Ren X, Luo M, Liu Y, Liu P, et al. Characterization of intratumor microbiome in cancer immunotherapy. The Innovation. 2023;4(5).

103. Elias EE, Lyons B, Muruve DA. Gasdermins and pyroptosis in the kidney. Nature Reviews Nephrology. 2023;19(5):337-50.

104. Wang H, Zhou X, Li C, Yan S, Feng C, He J, et al. The emerging role of pyroptosis in pediatric cancers: from mechanism to therapy. Journal of Hematology & Oncology. 2022;15(1):140.

105. Liu Y, Lu Y, Ning B, Su X, Yang B, Dong H, et al. Intravenous Delivery of Living Listeria monocytogenes Elicits Gasdmermin-Dependent Tumor Pyroptosis and Motivates Anti-

View Article Online
DOI: 10.1039/D5PM00045A

Tumor Immune Response. ACS Nano. 2022;16(3):4102-15.

106. Lou X, Wang J, Jin X, Wang X, Qin B, Liu D, et al. An oral bacterial pyroptosis amplifier against malignant colon cancer. Nano Today. 2024;54:102091.

107. Kalaora S, Nagler A, Nejman D, Alon M, Barbolin C, Barnea E, et al. Identification of bacteria-derived HLA-bound peptides in melanoma. Nature. 2021;592(7852):138-43.

108. Naghavian R, Faigle W, Oldrati P, Wang J, Toussaint NC, Qiu Y, et al. Microbial peptides activate tumour-infiltrating lymphocytes in glioblastoma. Nature. 2023;617(7962):807-17.

109. Klobuch S, Seijkens TTP, Schumacher TN, Haanen JBAG. Tumour-infiltrating lymphocyte therapy for patients with advanced-stage melanoma. Nature Reviews Clinical Oncology. 2024;21(3):173-84.

110. Backlund C, Jalili-Firoozinezhad S, Kim B, Irvine DJ. Biomaterials-Mediated Engineering of the Immune System. Annual Review of Immunology. 2023;41(1):153-79.

111. Ma X, Li S-J, Liu Y, Zhang T, Xue P, Kang Y, et al. Bioengineered nanogels for cancer immunotherapy. Chemical Society Reviews. 2022;51(12):5136-74.

112. Boehnke N, Straehla JP, Safford HC, Kocak M, Rees MG, Ronan M, et al. Massively parallel pooled screening reveals genomic determinants of nanoparticle delivery. Science. 2022;377(6604).

113. Zhu X, Xu J, Ling G, Zhang P. Tunable metal–organic frameworks assist in catalyzing DNAzymes with amplification platforms for biomedical applications. Chemical Society Reviews. 2023;52(21):7549-78.

View Article Online
DOI: 10.1039/D5PM00045A

114. Han X, Alu A, Liu H, Shi Y, Wei X, Cai L, et al. Biomaterial-assisted biotherapy: A brief review of biomaterials used in drug delivery, vaccine development, gene therapy, and stem cell therapy. Bioactive Materials. 2022;17:29-48.

115. Pacheco C, Baião A, Ding T, Cui W, Sarmento B. Recent advances in long-acting drug delivery systems for anticancer drug. Advanced Drug Delivery Reviews. 2023;194.

116. Erfani A, Diaz AE, Doyle PS. Hydrogel-enabled, local administration and combinatorial delivery of immunotherapies for cancer treatment. Materials Today. 2023;65:227-43.

117. Yan X, Ma F, Chen Q, Gou X, Li X, Zhang L, et al. Construction of size-transformable supramolecular nano-platform against drug-resistant colorectal cancer caused by Fusobacterium nucleatum. Chemical Engineering Journal. 2022;450:137605.

118. Wang DY, Cao Y, Yang G, Zhang S, van der Mei HC, Ren Y, et al. Self-Targeted Co-Delivery of an Antibiotic and a Cancer-Chemotherapeutic from Synthetic Liposomes for the Treatment of Infected Tumors. Advanced Functional Materials. 2023;33(32).

119. Li X, Ma Y, Xin Y, Ma F, Gao H. Tumor-Targeting Nanoassembly for Enhanced Colorectal Cancer Therapy by Eliminating Intratumoral Fusobacterium nucleatum. ACS Applied Materials & Interfaces. 2023;15(11):14164-72.

120. Song W-F, Zheng D, Zeng S-M, Zeng X, Zhang X-Z. Targeting to Tumor-Harbored Bacteria for Precision Tumor Therapy. ACS Nano. 2022;16(10):17402-13.

121. Wang M, Rousseau B, Qiu K, Huang G, Zhang Y, Su H, et al. Killing tumor-associated bacteria with a liposomal antibiotic generates neoantigens that induce anti-tumor immune

View Article Online
DOI: 10.1039/D5PM00045A

responses. Nature Biotechnology. 2023.

122. Geng S, Guo P, Li X, Shi Y, Wang J, Cao M, et al. Biomimetic Nanovehicle-Enabled Targeted Depletion of Intratumoral Fusobacterium nucleatum Synergizes with PD-L1 Blockade against Breast Cancer. ACS Nano. 2024;18(12):8971-87.

123. Cao C, Zhang T, Yang N, Niu X, Zhou Z, Wang J, et al. POD Nanozyme optimized by charge separation engineering for light/pH activated bacteria catalytic/photodynamic therapy. Signal Transduction and Targeted Therapy. 2022;7(1):86.

124. Hou J, Xianyu Y. Tailoring the Surface and Composition of Nanozymes for Enhanced Bacterial Binding and Antibacterial Activity. Small. 2023;19(42).

125. Song N, Yu Y, Zhang Y, Wang Z, Guo Z, Zhang J, et al. Bioinspired Hierarchical Self-Assembled Nanozyme for Efficient Antibacterial Treatment. Advanced Materials. 2024;36(10):2210455.

126. Wang X, Chen Q, Zhu Y, Wang K, Chang Y, Wu X, et al. Destroying pathogen-tumor symbionts synergizing with catalytic therapy of colorectal cancer by biomimetic protein-supported single-atom nanozyme. Signal Transduction and Targeted Therapy. 2023;8(1).

127. Xi J, Wang Y, Gao X, Huang Y, Chen J, Chen Y, et al. Reverse intratumor bacteria-induced gemcitabine resistance with carbon nanozymes for enhanced tumor catalytic-chemo therapy. Nano Today. 2022;43:101395.

128. Qu X, Yin F, Pei M, Chen Q, Zhang Y, Lu S, et al. Modulation of Intratumoral Fusobacterium nucleatum to Enhance Sonodynamic Therapy for Colorectal Cancer with Reduced Phototoxic Skin Injury. ACS Nano. 2023;17(12):11466-80.

View Article Online
DOI: 10.1039/D5PM00045A

129. Chen L, Zhao R, Shen J, Liu N, Zheng Z, Miao Y, et al. Antibacterial Fusobacterium nucleatum-Mimicking Nanomedicine to Selectively Eliminate Tumor-Colonized Bacteria and Enhance Immunotherapy Against Colorectal Cancer. Advanced Materials. 2023;35(45).

130. Guo J, Zou Y, Huang L. Nano Delivery of Chemotherapeutic ICD Inducers for Tumor Immunotherapy. Small Methods. 2023;7(5):2201307.

131. Dalmasso G, Cougnoux A, Faïs T, Bonnin V, Mottet-Auselo B, Nguyen HTT, et al. Colibactin-producing

Escherichia coli

enhance resistance to chemotherapeutic drugs by promoting epithelial to mesenchymal transition and cancer stem cell emergence. Gut Microbes. 2024;16(1).

132. Jiang S-S, Xie Y-L, Xiao X-Y, Kang Z-R, Lin X-L, Zhang L, et al. Fusobacterium nucleatum-derived succinic acid induces tumor resistance to immunotherapy in colorectal cancer. Cell Host & Microbe. 2023;31(5):781-97.e9.

133. de Oliveira Alves N, Dalmasso G, Nikitina D, Vaysse A, Ruez R, Ledoux L, et al. The colibactin-producing

Escherichia coli

alters the tumor microenvironment to immunosuppressive lipid overload facilitating colorectal cancer progression and chemoresistance. Gut Microbes. 2024;16(1).

134. Kang X, Bu F, Feng W, Liu F, Yang X, Li H, et al. Dual-Cascade Responsive Nanoparticles Enhance Pancreatic Cancer Therapy by Eliminating Tumor - Resident Intracellular Bacteria. Advanced Materials. 2022;34(49).

View Article Online
DOI: 10.1039/D5PM00045A

135. Gao C, Wang X, Yang B, Yuan W, Huang W, Wu G, et al. Synergistic Target of Intratumoral Microbiome and Tumor by Metronidazole–Fluorouridine Nanoparticles. ACS Nano. 2023;17(8):7335-51.

136. Xie S, Wei L, Liu Y, Meng J, Cao W, Qiu B, et al. Size-tunable nanogels for cascaded release of metronidazole and chemotherapeutic agents to combat Fusobacterium nucleatum-infected colorectal cancer. Journal of Controlled Release. 2024;365:16-28.

137. Zhou S, Lin Y, Zhao Z, Lai Y, Lu M, Shao Z, et al. Targeted deprivation of methionine with engineered Salmonella leads to oncolysis and suppression of metastasis in broad types of animal tumor models. Cell Reports Medicine. 2023;4(6):101070.

138. Chen Z, Liu Y, Yu Y, Yang S, Feng J, Zhu Y, et al. Micro-to-Nano Oncolytic Microbial System Shifts from Tumor Killing to Tumor Draining Lymph Nodes Remolding for Enhanced Immunotherapy. Advanced Materials. 2024;36(7):2306488.

139. Liu R, Cao Z, Wang L, Wang X, Lin S, Wu F, et al. Multimodal oncolytic bacteria by coating with tumor cell derived nanoshells. Nano Today. 2022;45:101537.

140. Guo Y, Song M, Liu X, Chen Y, Xun Z, Sun Y, et al. Photodynamic therapy-improved oncolytic bacterial immunotherapy with FAP-encoding S. typhimurium. Journal of Controlled Release. 2022;351:860-71.

141. Goto Y, Iwata S, Miyahara M, Miyako E. Discovery of Intratumoral Oncolytic Bacteria Toward Targeted Anticancer Theranostics. Advanced Science. 2023;10(20).

142. Nakatsuji T, Chen TH, Butcher AM, Trzoss LL, Nam S-J, Shirakawa KT, et al. A commensal strain of Staphylococcus epidermidis protects against skin neoplasia.

View Article Online
DOI: 10.1039/D5PM00045A

Science Advances. 2018;4(2):eaao4502.

Data Availability Statement (DAS)

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No primary research results, software or code have been included and no new data were generated or analysed as part of this review (**PM-REV-02-2025-000045.R1**).