The microbiome and mycobiome in esophageal cancer: Implications for pathogenesis, detection and treatment

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Background and objective: Esophageal cancer (EC), including the histologic subtypes esophageal adenocarcinoma (EAC) and esophageal squamous cell carcinoma (ESCC), is a leading cause of cancer-related mortality worldwide. Despite advancements in therapy, including the use of immune checkpoint inhibitors, 5-year survival remains poor. This narrative article summarizes the emerging role of the microbiome and mycobiome as potentially modifiable contributors to esophageal carcinogenesis, disease progression and treatment response. **Methods:** For this narrative article, a literature search was performed using PubMed and ClinicalTrials.gov. The search strategy included the keywords 'microbiome,' 'mycobiome,' 'esophageal squamous cell carcinoma' or 'esophageal adenocarcinoma.' Findings were synthesized and presented descriptively. **Key content and findings:** Common patterns of dysbiosis, lower bacterial diversity and microbial-induced alterations in inflammatory and metabolic pathways characterize both EAC and ESCC. In EAC, progression from Barrett's esophagus to adenocarcinoma is associated with a shift from a gram-positive, *Streptococcus*-dominant microbiome toward gram-negative anaerobes, linked mechanistically to lipopolysaccharide–Toll-like receptor 4–nuclear factor κB inflammatory signaling and bile acid–microbiome interactions. In ESCC, *Fusobacterium nucleatum* and *Porphyromonas gingivalis* are enriched and correlate with advanced disease stage, chemotherapy resistance and impaired response to immunotherapy, representing potential biomarker candidates. Preliminary evidence suggests that fungal dysbiosis, mycotoxin production and fungal-mediated immune modulation may influence the development of EC and treatment response. Altering the microbiome through fecal microbiota transplantation and targeted antimicrobial strategies shows preliminary efficacy in overcoming immunotherapy resistance in early-phase trials, although EC-specific data remain limited. **Conclusions:** This narrative article summarizes current evidence on the influence of the microbiome and mycobiome on carcinogenesis, progression and therapeutic response in EC. Larger prospective studies and randomized trials are needed to validate microbial biomarkers and translate microbiome-directed therapies into clinical practice.

Keywords

Barrett esophagus, esophageal adenocarcinoma, esophageal neoplasms, esophageal squamous cell carcinoma, fecal microbiota transplantation, gastrointestinal microbiome, mycobiome

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Esophageal cancer (EC) is a major global health concern, ranking as the 11th most commonly diagnosed cancer and the seventh leading cause of cancer-related deaths worldwide, with an estimated 494,320 new cases and 441,994 deaths in 2024.[1] Esophageal squamous cell carcinoma (ESCC) remains the predominant subtype globally (84% of new cases), while esophageal adenocarcinoma (EAC) now accounts for the majority of cases in high-income Western countries. ESCC predominates in Eastern Asia and parts of Africa, whereas EAC has risen sharply in the USA, Australia, Canada and several countries in Western and Northern Europe.[2] These differences in geography and risk factor exposure suggest that ESCC and EAC are not simply histologic variants of one disease but biologically distinct malignancies shaped by different environmental exposures, host factors and local tissue microenvironments.

Despite improvements in multimodality therapy, outcomes for EC remain poor. According to U.S. Surveillance, Epidemiology and End Results program data, the overall 5-year relative survival rate for EC is 22.2%, while the 5-year survival rate is 5.3% for distant disease. The near convergence of global incidence and mortality underscores how frequently EC remains lethal. These figures underscore the need for earlier detection, better risk stratification and a more precise understanding of underlying pathophysiology.[3]

The two histologies also differ in anatomy and pathophysiology. ESCC arises in the upper or mid esophagus and, similar to other aerodigestive

malignancies, is linked to tobacco use, alcohol consumption, dietary carcinogens and chronic mucosal injury.[4] EAC develops in the distal esophagus or gastroesophageal junction (GEJ) due to chronic gastroesophageal reflux disease, associated with central obesity, Barrett's esophagus (BE) and male sex. BE is the only established precursor lesion for EAC and represents the metaplastic consequence of long-standing reflux injury. EAC is therefore a particularly useful model for microbiome research because its pre-malignant progression sequence is endoscopically visible and accessible to biopsy. In certain patients, reflux esophagitis progresses to Barrett's metaplasia, followed by dysplasia and ultimately invasive adenocarcinoma. This stepwise progression provides an opportunity to assess whether microbial changes merely reflect epithelial transformation or actively participate in it. These differences between the two histologies matter clinically and therapeutically because the mechanisms driving initiation, evolution and treatment resistance are unlikely to be identical across subtypes, and any microbiome-based model of esophageal carcinogenesis must be interpreted within a histology-specific framework.[5] The EAC microbiome literature is less mature than that for ESCC but is most comprehensive around the BE–dysplasia–EAC sequence, where tissue, brushing, saliva and multi-omics studies now support an association between microbial community shifts and neoplastic progression.[6] The microbiome has emerged as a promising line of investigation in this space because it is dynamic, measurable, mechanistically active and potentially modifiable. Microbial signatures could help refine cancer risk assessment, identify biologically relevant disease states and inform prevention or therapeutic strategies, as summarized in the graphical abstract (*Figure 1*).

Methods

For this narrative article, a literature search was performed using PubMed and ClinicalTrials.gov. The search strategy included the keywords 'microbiome,' 'mycobiome,' 'esophageal squamous cell carcinoma' or 'esophageal adenocarcinoma,' yielding 334 results. Studies published between January 1, 1997, and May 15, 2026, were evaluated. Studies were independently analyzed by two reviewers, and disagreements on inclusion were resolved through discussion. Peer-reviewed, English-language studies that included adult patients (≥18 years) with either EAC or ESCC were considered. Sixteen studies evaluating microbiome or mycobiome profiles using human tissue or biospecimens from adult patients were included in the primary narrative synthesis (*Tables 1 and 2*).[7–22] Meta-analyses, systematic reviews, preclinical studies and relevant conference abstracts were additionally incorporated to provide context and further explore potential mechanisms of disease. Findings were synthesized and presented descriptively.

Microbiome and esophageal adenocarcinoma

Dysbiosis in esophageal metaplasia and dysplasia

The traditional view of the esophagus as a nearly sterile conduit was replaced by a model in which the normal esophagus harbors a reproducible microbial community resembling the oral microbiome but with its own mucosa-associated features. Early bacterial ribosomal RNA (16S rRNA) gene-based studies identified a 'type I' esophageal microbiome dominated by gram-positive organisms, particularly *Streptococcus*, in histologically normal tissue. In contrast, reflux esophagitis and BE were more often associated with

Figure 1: Graphical abstract

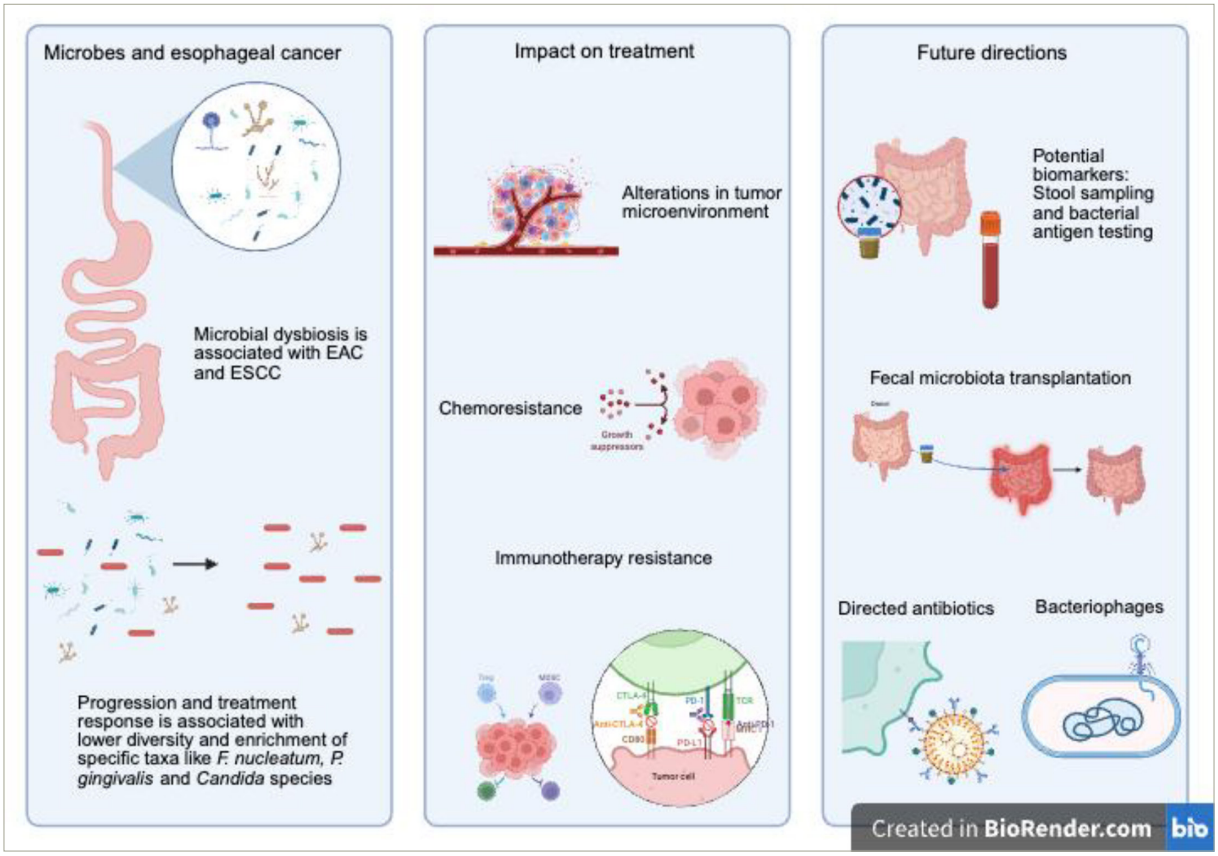


Figure created in BioRender.com (Toronto, ON, Canada)
EAC = esophageal adenocarcinoma; ESCC = esophageal squamous cell carcinoma; F. nucleatum = Fusobacterium nucleatum; = P. gingivalis = Porphyromonas gingivalis*; CTLA-4 = cytotoxic T-lymphocyte-associated protein 4, PD-1 = programmed cell death protein 1; PD-L1 = programmed cell death-ligand 1, TCR = T-cell receptor.*

Table 1: Microbiome impact on response to therapy in esophageal squamous cell carcinoma and esophageal adenocarcinoma[7–18]

Study	Participants	Therapy	Sampling	Microbial identification	Results
Chemotherapy response					
Ohsawa et al., 2026 Japan [8]	ESCC (n=74)	Cisplatin/5-FU, nedaplatin/5-FU or docetaxel/cisplatin/5-FU ± radiation followed by esophagectomy	Oral tongue coating	16S rRNA (V1–V2)	Alpha diversity correlated with pathologic response (OR, 11.46; 95% CI, 2.49–52.64; p=0.002), RFS (HR, 3.89; 95% CI, 1.57–9.65; p=0.005) and OS (p=0.044) Poor responders had higher abundance of *Streptococcaceae* and *Corynebacteriaceae*
Wu et al., 2024 China [9]	ESCC (n=180); 48 paired tissue samples (pre- and post-neoadjuvant chemoradiotherapy) from 24 patients	Platinum combined with paclitaxel chemotherapy + radiotherapy	Esophagectomy tissue samples	16S rRNA (V3–V4)	A microbiota-based risk prediction model on the genera of *TM7x*, *Sphingobacterium* and *Prevotella* was predictive of disease-free survival (p=0.00074 in internal validation set and p=0.0017 in independent validation set) and OS (p=0.00023 in internal validation set and p=0.11 in independent validation set)
Li et al., 2024 China [10]	ESCC (n =31)	Paclitaxel and cisplatin	Fecal samples pre- and post-treatment	16S rRNA (V4)	*Bacteroides plebeius* was more abundant in baseline samples from responders than non-responders (p=0.045)
Yamamura et al., 2019 Japan [11]	ESCC (n=551); 101 of those patients had received neoadjuvant chemotherapy	Training cohort: surgical resection alone (n=394) Validation cohort: neoadjuvant chemoradiation (n=41), neoadjuvant chemotherapy (n=101) with DCF regimen, neoadjuvant chemotherapy unspecified regimen (n=15)	Esophagectomy surgical samples	qPCR	Higher abundance of *Fusobacterium nucleatum* was associated with poor RFS in both cohorts (training cohort HR=1.61; p=0.03, validation cohort HR=1.96; p=0.004) Higher abundance of *F. nucleatum* was associated with worse chemotherapeutic response for all evaluation methods: RECIST (p=0.04), SUV_{max} change in PET/CT (p=0.0004) and TRG (p=0.003)
Liu et al., 2021 Japan [12]	ESCC (n=120 post-treatment samples, n=30 pre-treatment samples)	Neoadjuvant cisplatin and 5-FU ± docetaxel followed by surgical resection	Esophagectomy surgical samples for post-treatment; endoscopic biopsy for pre-treatment	qPCR	Fewer responders to chemotherapy in patients with high *F. nucleatum* burden compared with low burden (37.0% (10/27) versus 88.2% (60/68) in the low *F. nucleatum* group; p<0.001)
Yoshikawa et al., 2025 Japan [13]	ESCC (n=305)	Neoadjuvant CF or DCF or CRT with CF or DCF	Serum IgG-*Fn*	IgG-*Fn*	Positive group had significantly lower rates of OS and CSS as compared with patients who tested negative (OS; p=0.01, CSS; p=0.02) IgG-*Fn*-positive group showed a higher rate of pathologic progression and inadequate response to neoadjuvant treatment compared with the IgG-*Fn*-negative group (OR, 0.48; 95% CI, 0.24–0.92; p=0.04)
Immunotherapy response					
Xu et al., 2022 China [14]	ESCC (n=46)	Camrelizumab combined + carboplatin and albumin paclitaxel prior to surgical resection	Fecal samples	16S rRNA (V3–V4)	Alpha diversity declined after neoadjuvant chemoimmunotherapy Patients exhibiting pCR had different species and proportions of bacteria in pre-treatment samples compared with non-responders, although alpha diversity was not significantly different
Zhou et al., 2026 China [15]	ESCC (n=43); 122 fecal samples	Neoadjuvant cisplatin + paclitaxel; immunotherapy included pembrolizumab, sintilimab, toripalimab, camrelizumab or adrebelimab	Fecal samples	16S rRNA	Alpha diversity was higher in the responder group (p<0.05) *Ligilactobacillus salivarius* was enriched in non-responders (p<0.05)
Li et al., 2023 China [7]	ESCC (n=98)	PD-1 inhibitor	Serum IgG-*Fn*, surgical resection samples	IgG-*Fn*, qPCR	Patients with progressive disease had higher serum anti-*Fn* IgG levels and *Fn* DNA compared with partial responders (p=0.003) and those with stable disease (p=0.0045)
Wu et al., 2023 China [16]	ESCC; received neoadjuvant chemoimmunotherapy (n=25); received surgery without neoadjuvant therapy (n=15)	Neoadjuvant chemoimmunotherapy (exact regimen unspecified), PD-1 inhibitor	Surgical resection samples, fecal samples	qRT-PCR	Abundance of *Streptococcus* positively correlated with disease-free survival (p=0.034) and $GrzB^+$ and $CD8^+$ T-cell infiltration in tumor tissues
Van den Ende et al., 2024 Netherlands [17]	n=172 with resectable esophageal carcinoma (EAC + ESCC); 132 received CROSS alone, 40 received CROSS + anti-PD-L1. Mixed histology; CRT-only arm included ESCC; ICI arm was EAC/GEJ-dominant	Neoadjuvant CROSS (carboplatin + paclitaxel + 41.4 Gy) ± anti-PD-L1 (atezolizumab), followed by esophagectomy	Fecal samples at baseline, week 3 and pre-surgery; duodenal biopsies in the ICI arm	16S rRNA (V3–V4)	Distinct longitudinal fecal microbiome trajectories in responders versus non-responders; specific Lachnospiraceae taxa associated with pathologic response; intratumoral microbiota also shifted during treatment

Continued

Table 1: Continued

Study	Participants	Therapy	Sampling	Microbial identification	Results
Shaikh et al., 2024 USA [18]	Pilot cohort of patients with stage II–III resectable esophageal or GEJ carcinoma (EAC/GEJ)	Neoadjuvant ICI (anti-PD-1/PD-L1) combined with chemoradiation, prior to surgical resection	Paired fecal and tumor samples before and during treatment; concurrent fecal metabolomics	16S rRNA + untargeted fecal metabolomics	Patients achieving pCR showed enrichment of sphingolipid and primary bile-acid pathways with elevated *Roseburia inulinivorans*, *Ruminococcus callidus* and *Fusicatenibacter saccharivorans*; non-pCR patients lacked these SCFA-producing taxa

CF = cisplatin/5-fluorouracil; CI = confidence interval; CROSS = ChemoRadiotherapy for Oesophageal cancer followed by Surgery Study, refers to regimen of chemoradiotherapy with carboplatin and paclitaxel; CRT = chemoradiotherapy; CSS = cancer-specific survival; DCF = docetaxel, cisplatin and 5-fluorouracil; EAC = esophageal adenocarcinoma; ESCC = esophageal squamous cell carcinoma; F. nucleatum = Fusobacterium nucleatum*; 5-FU = 5-fluorouracil; GEJ = gastroesophageal junction; GrzB+ = granzyme B serine protease; HR = hazard ratio; ICI = immune checkpoint inhibitor; IgG-*Fn = *IgG antibody titer level against* Fusobacterium nucleatum*; OR = odds ratio; OS = overall survival; pCR = pathologic complete response ; PD-1 = programmed cell death protein 1; PD-L1 = programmed death-ligand 1; PET/CT = positron emission tomography–computed tomography; qPCR = quantitative polymerase chain reaction; qRT-PCR = quantitative reverse transcription polymerase chain reaction; RECIST = Response Evaluation Criteria in Solid Tumors; RFS = recurrence-free survival; SCFA = short-chain fatty acid; 16S rRNA = bacterial ribosomal RNA; SUV_{max} = standardized uptake value; TRG = tumor regression grade.*

a 'type II' microbiome enriched for gram-negative anaerobes and microaerophiles. This conceptual shift is important because it links microbial composition to disease state rather than simply to luminal contamination. It also provided one of the earliest frameworks for understanding how microbial populations might track the reflux–metaplasia axis that precedes EAC.[23]

Subsequent studies examining the BE–EAC sequence have supported the presence of dysbiosis, although the specific taxa associated with progression vary. Cross-sectional studies sampling controls, BE, dysplasia and EAC have shown that the microbiome changes as BE/pre-neoplasia advances, with reduced diversity in EAC compared with the *Firmicutes*-enriched pattern in healthy tissues. Other cohorts identified significant compositional differences among control mucosa, BE, dysplastic BE and adenocarcinoma, reinforcing the idea that microbial composition changes during epithelial transformation rather than only at the endpoint of invasive cancer.[6]

Although no single taxonomic signature is replicated across every cohort, several recurrent themes exist related in the progression from BE to EAC. Relative loss of *Streptococcus* and other gram-positive commensals and gain of gram-negative or oropharyngeal-associated taxa occur. In one cohort, *Streptococcus* and *Granulicatella* were less abundant in EAC mucosa than in BE mucosa, whereas other studies reported enrichment of taxa such as *Veillonella*, members of the *Enterobacteriaceae*, *Akkermansia* and *Lactobacillus fermentum* in EAC samples. The overall process reflects the development of EAC in the setting of a community-level departure from the *Streptococcus*-dominant pattern seen in the healthy distal esophagus.[24–26]

A multi-omics study by Deshpande et al. showed that the progression of BE is associated not only with community-level shifts but also with strain-level microbial signatures that are linked to host transcriptional changes. Enrichment of *Campylobacter* species was a progression-relevant signal that demonstrated that the esophageal microbiome occupies an ecological niche that is independent of the oropharynx.[27]

Potential mechanisms for dysbiosis-mediated carcinogenesis

The current mechanistic hypothesis linking dysbiosis to EAC centers on the chronic activation of innate immunity. The BE-related shift from a gram-positive, *Streptococcus*-dominant community toward gram-negative organisms may be due to increased local exposure to bacterial lipopolysaccharide (LPS). Translational research focuses on the LPS–toll-like receptor 4 (TLR4)–nuclear factor κB (NF-κB axis). Activation of TLR4 in BE is linked to NF-κB signaling activation and interleukin-8 (IL-8) induction. This provides a plausible route by which dysbiosis could sustain low-grade inflammation, epithelial injury and pro-neoplastic signaling over time. In this model, the microbiome is not just a biomarker of reflux injury but a biologically active amplifier.[28]

Campylobacter concisus is a candidate organism for activating this process. It is present in BE and is proposed as a progression-relevant organism. Mechanistic studies demonstrate a functional role rather than incidental colonization. Exposure of BE cell lines to *C. concisus* alters the expression of IL-18, tumor necrosis factor alpha (TNF-α) and p53 and upregulates programmed death-ligand 1 messenger RNA (PD-L1 mRNA), following *C. concisus* exposure.[29] This suggests that specific microbes influence inflammatory and immune-regulatory programs relevant to carcinogenesis.

A second mechanism involves the interaction between reflux chemistry, microbial metabolism and host epithelial signaling. Exposure to refluxed bile acids is a known cause of BE; however, newer studies suggest a causative interaction between bile acids and the microbiome. Increased refluxed bile acids correlate with BE-associated microbial configurations and host gene expression programs potentially relevant to EAC development. Complementary experimental work showed that microbiota-metabolized bile acids accelerate EAC through a diet–microbiome–metabolome axis.[30] These findings are still early and require further validation, but they point toward functional microbial metabolites as potential mediators of risk.

These findings support a working model in which EAC-associated dysbiosis contributes to carcinogenesis through three overlapping processes: the maintenance of chronic mucosal inflammation, modulation of epithelial stress and immune signaling and alteration of metabolite exposure within the distal esophageal niche.[28] Although most evidence remains associative, findings thus far suggest that microbial shifts may both influence and reflect carcinogenic change (*Figure 2*).

There are several limitations to these proposed mechanisms. First, most studies are cross-sectional and based on small cohorts. Second, low-biomass sampling creates substantial susceptibility to contamination, batch effects and platform-dependent differences. Third, studies vary widely in source material, including saliva, oral swabs, esophageal brushings, mucosal biopsies, resection specimens and stool; one recent study showed that tissue source itself can influence the observed esophageal flora.[26] Fourth, treatment and exposure confounders, such as proton pump inhibitors, antibiotics, reflux severity and diet, are difficult to standardize and may alter the very microbial signatures under study.[31]

Figure 2: Impact of microbiome on esophageal dysplasia

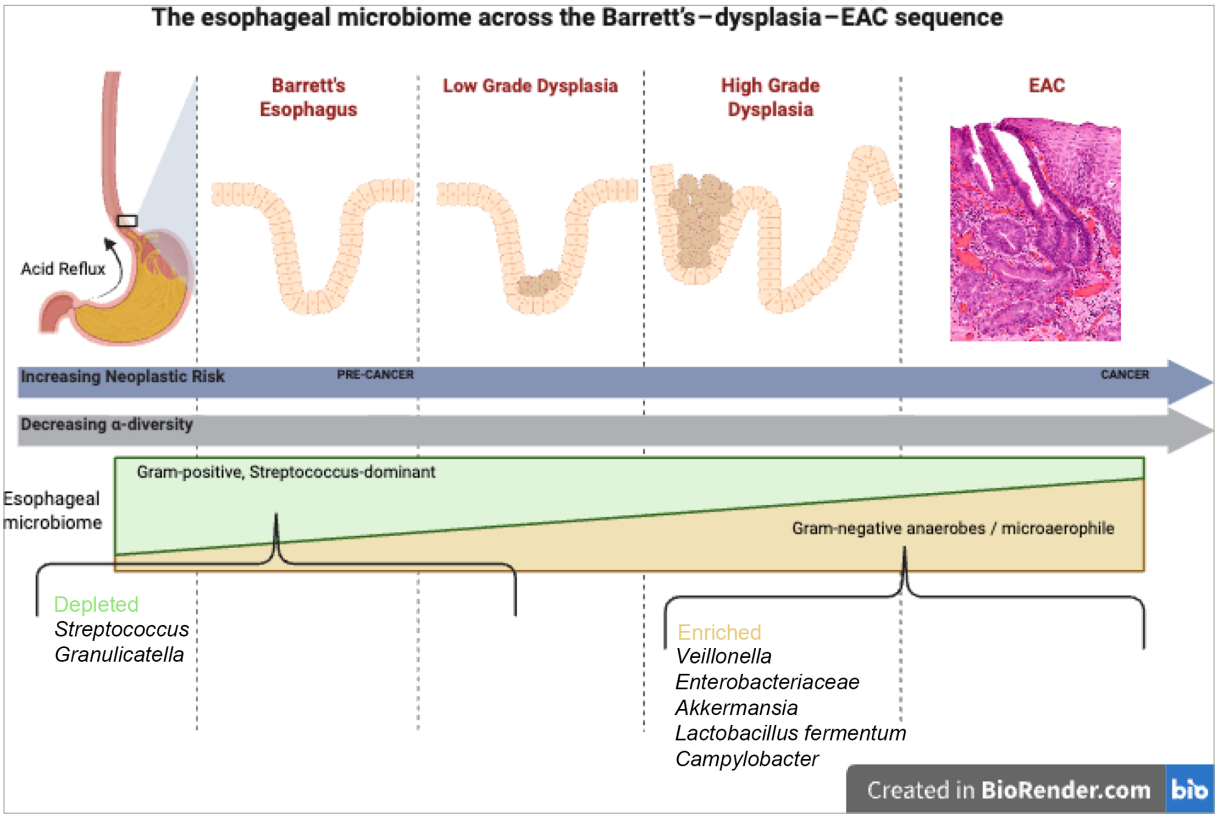


Figure created in BioRender.com (Toronto, ON, Canada)
EAC = esophageal adenocarcinoma.

These limitations help explain why the EAC literature often converges at the level of broad ecological change but diverges at the level of individual taxa. For the field to move forward, longitudinal studies that start in non-dysplastic BE, use standardized sampling and contamination controls, incorporate metabolomic or transcriptomic correlates and follow patients through clinically meaningful endpoints will be essential. Until then, the microbiome should be considered a biologically credible and clinically promising component of EAC pathogenesis, but not yet a validated stand-alone biomarker or therapeutic target.

Microbiome and esophageal squamous cell carcinoma

Intestinal dysbiosis in esophageal squamous cell carcinoma

There is a comparatively more robust body of literature on microbiome alterations and dysfunction in ESCC. Similar to BE and EAC, there is histological progression in ESCC starting with dysplasia. Fecal samples demonstrate dysbiosis of the intestinal microbiome in patients with ESCC compared with healthy controls, as well as common patterns of bacterial abundance across cohorts. *Fusobacterium*, *Streptococcus* and *Bacteroides* are typically more prevalent.[32–40] In contrast, taxa that have been associated with gut homeostasis are often depleted, such as *Prevotella*, *Lachnospira*, *Butyricicoccus* and *Faecalibacterium*.[33,35–37,40,41]

There are notable trends where patients with ESCC tend to have lower levels of bacteria that produce short-chain fatty acids (SCFAs), including butyrate.[36,37] There is a well-established link between butyrate depletion and gut barrier disruption via decreased expression of occludin and claudin-1, destabilization of hypoxia-inducible factor 1 and promotion of IL-22.[42] This yields a plausible mechanism for increased mucosal permeability, inflammation and bacterial translocation, although it has not been directly validated in ESCC. There is also an enrichment of LPS-producing bacteria in ESCC.[36,43] Notably, serum levels of LPS have been found to be elevated in patients with ESCC.[44] As a natural agonist of TLR4, LPS may activate pro-inflammatory signaling cascades (TNF-α, transforming growth factor-beta [TGF-β] and inhibition of IL-10) and cell proliferation (NF-kB, extracellular signal-regulated kinase and p38 mitogen-activated protein kinase).[45]

Dysregulated biosynthetic pathways may also contribute. Reduced levels of L-aspartate and pantothenic acid, with subsequent downregulation of the pantothenate and coenzyme A biosynthesis pathway, correlate with an increased abundance of *Enterobacteriaceae* and *Lactobacillus*.[34] One limitation of fecal sampling is its indirect representation of the esophageal tumor microenvironment, as it is more reflective of the intestinal microbiome than the microbiome within or adjacent to tumor tissue. However, there are clear practical advantages of fecal sampling, including its non-invasive nature and feasibility of longitudinal sampling, making it an important target for further research.

Esophageal dysbiosis in esophageal squamous cell carcinoma

In ESCC, the pathogenic bacteria of the genus *Fusobacterium* are extensively studied. *Fusobacterium nucleatum* (*Fn*), a gram-negative anaerobic bacterium commonly located in the oropharynx, colonizes the entire gastrointestinal tract, including the esophagus.[46,47] Multiple studies consistently demonstrate higher levels of *F. nucleatum* in ESCC compared with healthy esophageal mucosa.[48–53] *F. nucleatum* is implicated in ESCC progression, with its abundance correlating with the progression of clinical stage and poorer prognosis.[50,52,54–56]

Given the high association of *F. nucleatum* with the pathogenesis and progression of ESCC, multiple mechanisms are proposed to explain carcinogenesis. *F. nucleatum* is a facultative intracellular bacterium that can invade ESCC cells.[7,56] Methyltransferase-like protein 3 transcription is upregulated during intracellular infection, resulting in increased methylation and stabilization of the mRNA for oncogene c-Myc, a transcription factor, which may contribute to ESCC metastasis.[57] Intracellular *F. nucleatum* also activates the immune receptor nucleotide-binding oligomerization domain-containing protein 1 (NOD1), promoting the pro-inflammatory NOD1/receptor-interacting protein kinase 2 (RIPK2)/NF-κB pathway, thus providing a plausible link to uncontrolled cell proliferation, resistance to apoptosis and induction of the epithelial–mesenchymal transition (EMT), thereby enabling metastasis.[58,59] It also upregulates IL-32/proteinase 3, subsequently inducing the highly oncogenic phosphoinositide 3-kinase (PI3K)/protein kinase B (Akt) and aryl hydrocarbon receptor (AHR)/cytochrome P450 monooxygenase (CYP1A1)/Akt signaling pathways.[55,60]

A similar pattern emerges with other oral bacteria. *P. gingivalis,* which is implicated in periodontal disease, is also associated with ESCC.[61] Similar to *F. nucleatum,* it is enriched in tumor tissue and correlates with disease progression and poor prognosis.[62–64] Numerous potential mechanisms for this interaction are known, including activation of TGF-β/Smad signaling, enrichment of the PI3K/Akt pathway, inhibition of apoptosis via decreased purinergic receptor 7 (P2X7), promotion of the EMT through IL-6 activation, resistance to apoptosis dependent on fimbriae and signal transducer and activator of transcription 3 and TLR4/myeloid differentiation primary response 88 (MYD88)/c-Jun N-terminal kinase signaling via LPS, in addition to others.[64–74] Although *F. nucleatum* and *P. gingivalis* are the most extensively characterized, other taxa may also be associated with ESCC, including *Fusobacterium periodonticum*, streptococcal species, *Prevotella* and *Campylobacter*.[75–80] Although their role mechanistically is less defined, recurrent identification of these taxa suggests a broader microbial network that contributes to tumorigenesis and progression.

When considering the influence of the local esophageal microbiome on host metabolic pathways, specifically aldehyde metabolism, a mixed picture appears. Acetaldehyde, a byproduct of ethanol breakdown, is an established carcinogen and driver of ESCC development. Deficiency of the enzyme responsible for elimination of acetaldehyde, aldehyde dehydrogenase 2, increases the risk of ESCC in individuals who consume alcohol.[81] The local effects of acetaldehyde, including DNA damage and increased oxidative stress in tissues exposed to alcohol, have been proposed to mediate carcinogenesis of upper gastrointestinal tract cancers.[82] Several common oropharyngeal bacteria can produce significant quantities of acetaldehyde when exposed to ethanol, including *Neisseria, Streptococcus*, *Rothia* and *Prevotella*. Notably, these species are enriched in healthy mucosa, with only certain *Streptococcus* species inconsistently enriched in ESCC tumor tissue.[19,83] *F. nucleatum* and *Porphyromonas* do not produce acetaldehyde in significant quantities.[84] These expression patterns suggest that acetaldehyde-producing microbial populations may play a greater role in initial carcinogenesis than in tumor progression. Further studies are needed to elucidate the exact nature of this relationship, as numerous factors, such as environmental exposures, genetics and behavioral patterns of alcohol consumption, all influence this metabolic pathway.[85]

Influence of the microbiome on response to therapeutics in esophageal cancer

Chemotherapy resistance patterns

Beyond the role of the microbiome in ESCC development, there is increasing evidence that microbial composition may influence pathologic response. Multiple studies have identified alterations in response to neoadjuvant chemotherapy based on patterns of microbial diversity and the abundance of certain bacterial taxa, particularly *F. nucleatum* (*Table 1*). Higher bacterial alpha diversity in oral samples correlates with pathologic response, recurrence-free survival and overall survival in patients treated with platinum/5-fluorouracil (FU)-based regimens. Responders had enrichment of *Lactobacillales*, *Peptostreptococcales–Tissierellales*, *Bifidobacteriaceae*, *Erysipelotrichaceae*, *Lactobacillaceae*, *Anaerovoracaceae*, *Staphylococcaceae* and *Aerococcaceae,* whereas *Streptococcaceae* and *Corynebacteriaceae* were enriched in non-responders.[8] These flora are derived from the oral microbiome rather than from direct sampling of the esophageal mucosa but may be anatomically relevant, as oral microbial species can move to the esophageal and gastrointestinal tracts via microaspiration, reflux or bacteremia.[86] Although further investigation is needed to clarify the relationship between the oral microbiome and esophageal pathology, oral microbial sampling could represent a minimally invasive and highly accessible method for predicting treatment response. It also raises intriguing questions regarding the potential role of low-cost interventions aimed at improving oral health in enhancing treatment response.[87,88]

Multiple pre-treatment bacterial taxa are associated with either enhanced or impaired therapeutic response. A microbiota-based risk prediction model incorporating the genera *TM7x*, *Sphingobacterium* and *Prevotella* in esophageal tissue samples predicted disease-free survival (DFS) in patients treated with cisplatin-based chemotherapy combined with radiotherapy.[9] Similarly, fecal microbiome analyses have shown that the abundance of *Bacteroides plebeius* correlates with chemotherapy response to cisplatin/paclitaxel, suggesting a protective or sensitizing effect of certain microbial populations.[10] Conversely, *P. gingivalis* is associated with high expression of the inflammatory protein glycogen synthase kinase-3 beta (GSK3β) in ESCC cells, which could induce chemoresistance and disease progression via GSK3β-mediated mitochondrial oxidative phosphorylation.[89,90]

As previously discussed, *F. nucleatum* is implicated in ESCC development through multiple mechanisms.[55,56,91] It may also influence the response to chemotherapy. Treatment with docetaxel, cisplatin and 5-FU (DCF) in patients with a higher abundance of *F. nucleatum* in esophagectomy specimens is associated with advanced clinical stage, poor recurrence-free survival and reduced therapeutic response, as measured by multiple modalities (standard uptake value, positron emission tomography/computed tomography and pathologic tumor regression grade).[11] Similar findings were observed in patients treated with cisplatin/5-FU (CF) ± docetaxel who had lower response rates with a high *F. nucleatum* burden.[12] As an alternative sampling modality, serum immunoglobulin G (IgG) antibody titer levels against *F. nucleatum* (IgG-*Fn*) predict the response to neoadjuvant chemotherapy (CF and DCF regimens with or without radiotherapy). Positive IgG-*Fn* correlates with poor cancer-specific and overall survival.[13] Although further validation is necessary, a serum *F. nucleatum* antigen test may be a non-invasive biomarker for risk stratification and treatment planning. Notably, an IgG test would not

differentiate current colonization from past exposure to *F. nucleatum* in a given individual.[92]

The mechanisms by which *F. nucleatum* impacts therapeutic response are not fully understood. Bacterium-induced drug resistance occurs in colorectal cancer models via TLR4 and MYD88 signaling and microRNA (miRNA), a class of non-coding RNA production, as well as in pancreatic cancer via the conversion of gemcitabine into an inactive metabolite by Gammaproteobacteria.[93,94] *In vitro* and *in vivo* models demonstrate that high NOD-like receptor protein 3 expression and the presence of myeloid-derived suppressor cells in ESCC are associated with a decline in cisplatin sensitivity, suggesting an immunomodulatory component of chemoresistance.[95] Autophagy may also contribute to resistance, given that *F. nucleatum* induces autophagosome formation and increases *Autophagy related 7 gene* (*ATG7*) and Beclin 1 levels in ESCC cells. Inhibition of autophagy via *ATG7* knockdown reversed chemoresistance.[12] *F. nucleatum* invades chemotherapy-induced senescent ESCC cells, stimulating paracrine signaling via the senescence-associated secretory phenotype promoting progression, recurrence and chemoresistance.[96]

Immunotherapy resistance patterns

Since the publication of landmark studies that established the efficacy of chemoimmunotherapy (CIT) and its increasing incorporation into clinical practice, there are on-going attempts to characterize the impact of the microbiome on immunotherapy response.[97–102] Poor responders to CIT demonstrate enrichment of distinct bacterial taxa in pre-treatment fecal samples compared with patients who achieve a pathologic complete response. Exposure to neoadjuvant CIT reduces alpha diversity in tumor samples.[14] This contrasts with findings of improved response to chemotherapy in individuals with higher alpha diversity in pre-treatment specimens and with findings in immunotherapy responders.[8,91] This suggests a complex and potentially bidirectional relationship in which pre-treatment microbial diversity influences response, while subsequent treatment with CIT may produce treatment-favorable microbial populations. Supporting this concept, chemotherapy disrupts the microbiome independently of antibiotic exposure, indicating that treatment-related microbial alterations may be due to a direct biological effect rather than a consequence of supportive care measures.[103]

Specific bacterial strains are involved in the immunotherapy response, particularly *F. nucleatum*. In patients treated with a programmed cell death protein 1 (PD-1) inhibitor, both higher levels of *F. nucleatum* DNA in tumor tissue samples and serum *Fn* antibodies were identified in non-responders. Proposed mechanisms for *F. nucleatum* interference in immunotherapy include inhibition of T-cell cytokine secretion and binding of the virulence factor Fn-DNA hunger/stationary phase protective protein to the *PD-L1* gene promoter. This upregulates PD-L1 expression via activation of transcription factor 3, allowing tumor cells to evade the T-cell-mediated immune responses.[7] Alternatively, *F. nucleatum* may promote immune evasion and tumor growth through stromal remodeling. *F. nucleatum* activates the NF-κB pathway in tumor cells via NOD1/RIPK2.[58] CD8+ T-cell infiltration is subsequently inhibited, and there is an increase in cancer-associated fibroblasts within the tumor microenvironment, providing a plausible mechanism for the suppression of PD-1 inhibitor activity.[91]

Additional bacterial taxa modulate PD-1 inhibition. *Ligilactobacillus salivarius* is amplified in non-responders. *In vitro* and *in vivo* models have demonstrated that indole-3-lactic acid, produced by *L. salivarius,* suppresses NKG7-expressing CD8+ Tpex (precursor exhausted T) cells within tumor tissue, thereby exacerbating PD-1 resistance.[15] Conversely, certain bacterial populations may enhance immunotherapeutic efficacy. Higher levels of *Streptococcus* correlate with longer DFS , granzyme B serine protease and CD8+ T-cell infiltration of tumor tissue. In murine models, fecal microbiota transplantation led to higher *Streptococcus* levels in tumor tissue associated with increased CD8+ T-cell infiltration and an improved response to PD-1 inhibition.[16] C chemokine receptor 1 (CCR1) is implicated in T-cell exhaustion and poor response to PD-1 inhibition.[104] Bacterial LPS, a metabolite of gram-negative bacilli, upregulates the expression of both CCR1 and PD-L1 in mouse models. There is a predominance of gram-negative bacteria in the esophageal flora, supporting another explanation for poor response to immunotherapy.[43] Together, these data support the hypothesis that the microbiome modulates PD-1 inhibitor efficacy through multiple interconnected pathways, including mediation of T-cell function, production of metabolites and regulation of checkpoint-associated signaling. Despite advances in EAC treatment with CIT, few studies have examined the modulation of treatment response by the microbiome.[105] Similar to the patterns demonstrated in ESCC, fecal alpha diversity is higher in responders. Low taxonomic diversity and a high burden of *Fusobacterium* are associated with poor progression-free survival (PFS).[17] There are also alterations in metabolic profiles, with responders showing enrichment of sphingolipid and primary bile-acid pathways, along with elevated levels of *Roseburia inulinivorans*, *Ruminococcus callidus* and *Fusicatenibacter saccharivorans*, whereas non-responders lacked these SCFA-producing taxa.[18] Additional studies are needed to further characterize these complex relationships in EAC.

Emerging role of the fungal mycobiome

The role of the fungal mycobiome in ESCC is an emerging field of study (*Table 2*). Technological advancements have improved fungal profiling, which has led to a greater understanding of the crucial role the mycobiome plays in immune regulation in healthy individuals.[106–108] This occurs through the modulation of cytokine production, which influences T-cell responses, and interactions with pattern recognition receptors such as Dectin-1 and TLRs.[109–111] Importantly, fungal dysbiosis is implicated in other gastrointestinal malignancies, including pancreatic and colorectal cancers.[112,113] Although current ESCC literature is limited, early evidence suggests that similar patterns of fungal dysbiosis, lack of species diversity, mycotoxin production and modulation of immune response and metabolic pathways may influence development of ESCC.

In a study comparing esophageal biopsy samples from ESCC, esophageal squamous intraepithelial neoplasia (ESIN) and healthy controls, higher abundances of *Alternaria*, *Neosetophoma* and *Monascus* were demonstrated in the ESCC group.[19] *Neosetophoma* and *Monascus* were not previously implicated in carcinogenesis. *Alternaria alternata* is an emerging mycotoxin-producing fungus, which is linked to EC as the main fungal contaminant of grain in areas of China with high ESCC burden.[114,115] *Alternaria* produces alternariol monomethyl ether and alternariol, which induce transformation of mouse embryonic fibroblast cells, promote proliferation of human esophageal epithelium, interact with DNA topoisomerase and increase the production of reactive oxygen species *in vitro*.[116–118] Notably, *Aspergillus* was the key fungal genus identified in ESIN samples. *Aspergillus* has not been studied in EC, although it is associated with the suppression of cytotoxic T cells in lung adenocarcinoma through the activation of myeloid-derived

Table 2: Mycobiome in esophageal cancer[19–22]

Study, country	Participants	Sampling	Microbial identification	Results
Yuan et al., 2025 China [19]	HC (n=13), ESCC (n=12), ESIN (n=10)	Endoscopic biopsies (HC), biopsies of ESCC or ESIN lesions and adjacent tissue obtained in patients with ESCC and ESIN	ITS sequencing and 16S rRNA amplicon sequencing (V3–V4)	*Alternaria*, *Neosetophoma* and *Monascus* were more prevalent in the ESCC group, although not significantly different between samples with disease and adjacent tissue *Aspergillus* was dominant in the ESIN group and was enriched in samples with disease compared with adjacent tissue *Coniochaeta* and *Sclerostagonospora* were more prevalent in the HC group Tumor tissue had lower alpha diversity compared with adjacent tissue (p=0.001)
Rao et al., 2023 China [20]	ESCC (n=66), no HC	Paired tumor and tumor-adjacent surgical samples were obtained from patients with ESCC undergoing esophagectomy	ITS sequencing and 16S rRNA amplicon sequencing (V3–V4), qRT-PCR	*Rhodotorula toruloides*, *Malassezia dermatis*, *Hanseniaspora lachancei* and *Spegazzinia tessarthra* were more abundant in tumor samples *Preussia persica*, *Fusarium solani*, *Nigrospora oryzae*, *Acremonium furcatum*, *Golovinomyces artemisiae* and *Tausonia pullulans* were more abundant in tumor-adjacent samples Expression of MET proto-oncogene, receptor tyrosine kinase (MET) was positively correlated with *S. tessarthra*
Liang et al., 2025 China [21]	ESCC (n=68), HC (n=19)	Fecal samples	ITS sequencing	Baseline samples in ESCC showed reduced alpha diversity (p<0.001), with enrichment of pathogenic fungi *Rhodotorula minuta, Actinomucor elegans* and *Candida zeylanoides* Responders to neoadjuvant immunotherapy showed higher fungal diversity and enrichment of beneficial strains (*Candida boidinii*, *Meyerozyma guilliermondii* and *Trichosporon dermatis*), correlating with elevated cytotoxic markers Non-responders to immunotherapy showed enrichment of immunosuppressive fungi (*Candida albicans*, *Candida parapsilosis*, *Candida glabrata* and *Saccharomyces*), correlating with Th2 skewing and increased regulatory cytokines
Dohlman et al., USA [22]	Pan-cancer mycobiome reanalysis; EC n=142 primary tumors (mixed EAC + ESCC, not separated)	Primary tumor tissue (TCGA WGS reads)	PathSeq pipeline on TCGA WGS; prevalence-based decontamination; external ITS validation; culture-dependent confirmation	Esophageal tissue had lower fungal DNA abundance than colon/stomach/head-neck. GI tumors classifiable as *Candida*-dominant (Ca-type) or *Saccharomyces*-dominant (Sa-type). Multiple *Candida* species (*C. albicans, C. tropicalis, C. glabrata*) enriched in tumors versus adjacent normal; tumor-associated *Candida* DNA predictive of decreased survival across GI sites

EAC = esophageal adenocarcinoma; EC = esophageal cancer; ESCC = esophageal squamous cell carcinoma; ESIN = esophageal squamous intraepithelial neoplasia; GI = gastrointestinal; HC = healthy controls; ITS = internal transcribed spacer; MET = mesenchymal–epithelial transition proto-oncogene, receptor tyrosine kinase; qRT-PCR = quantitative reverse transcription polymerase chain reaction; 16S rRNA = bacterial ribosomal RNA; TCGA = The Cancer Genome Atlas; Th2 = subset of T helper cell involved in antibody-mediated immunity; WGS = whole-genome sequencing.

suppressor cells, mediated by IL-1β secretion via the β-glucan/Dectin-1/CARD9 pathway, which is involved in the recognition of fungal antigens by the innate immune system.[119] While direct evidence in esophageal malignancy is currently lacking, the identification of *Aspergillus* in pre-malignant lesions and other tumor types suggests that fungal-mediated immune modulation may drive malignant transformation and subsequent immune evasion.

Another study demonstrated higher levels of *Rhodotorula toruloides*, *Malassezia dermatis*, *Hanseniaspora lachancei* and *Spegazzinia tessarthra* in ESCC tumor samples compared with adjacent tissue in esophagectomy specimens.[20] The composition of fungal populations was associated with the geographic origin of patients, which is consistent with the known relationship between the environment and an individual's mycobiome.[120] One enriched species, *Malassezia,* is implicated in tumorigenesis in pancreatic ductal adenocarcinoma. One mechanistic driver in pancreatic cancer is activation of the complement cascade via mannose-binding lectin.[113] *R. toruloides* is a recognized opportunistic pathogen that is thought to be involved in multiple metabolic processes, such as fatty acid biosynthesis and glucose metabolism, although it is not yet implicated in oncogenesis.[121–124] Species richness and diversity were lower in tumor samples compared with adjacent tissue. Using co-expression analysis of biomarkers, the abundance of certain species correlated with the expression of genes *MTOR* and *MET*. Higher levels of expression were observed in tumor-adjacent samples with abundant *Nigrospora oryzae* and *Tausonia pullulans,* as well as in tumor samples with abundant *S. tessarthra.*[20] This raises the question of whether certain fungal strains may impact gene expression in both healthy tissue and the tumor microenvironment, influencing the development and progression of ESCC.

Despite the mycobiome representing a relatively new focus of study, efforts are underway to analyze whether it meaningfully impacts host response to immunotherapy. Liang et al. compared fecal samples from patients with ESCC with those from healthy controls. Baseline samples revealed a lack of alpha diversity and enrichment of pathogenic fungi in ESCC samples, specifically *Rhodotorula minuta, Actinomucor elegans* and *Candida zeylanoides*. These baseline profiles predicted response to immune checkpoint inhibitor (ICI) therapy, with responders exhibiting higher diversity and an abundance of beneficial taxa such as *Candida boidinii, Meyerozyma guilliermondii* and *Trichosporon dermatis*. Non-responders exhibited a higher abundance of immunosuppressive fungi (*Candida albicans, Candida parapsilosis, Candida glabrata, Saccharomyces*). Furthermore, responders had increased expression of cytotoxic markers (interferon-γ, IL-2) and greater Th1-polarized immunity compared with higher levels of regulatory cytokines (IL-4, IL-10, IL-13) and Th2 skew in non-responders. Th1 refers to type 1 helper T cells promoting cell-mediated immunity, while Th2 refers to type 2 helper T cells involved in antibody-mediated immunity. In this trial, participants were excluded if they had recent exposure to antibiotics or immunosuppressants within 4 weeks of sample collection. Participants selected as healthy controls were cohabitants of the patients with ESCC to control for variation in lifestyle or dietary habits. There are clear limitations in drawing broad conclusions on the association between fungal dysbiosis and immunotherapy response given the limited data available in this singular retrospective cohort study.[21] These initial findings merit further

investigation to characterize how fungal community composition may correlate with both treatment response and the broader immune milieu associated with effective antitumor immunity in different patient populations.

The overexpression of *Candida* species in immunotherapy non-responder samples is particularly notable, as these yeasts have also been associated with increased metastatic potential in other tumor types and may suppress cytotoxic T-cell activity, thereby promoting immune evasion.[125–128] It has also been linked with ESCC development, with one retrospective cohort study of 207 patients with achalasia identifying an association between prior esophageal *Candida* infection and development of EC (adjusted hazard ratio, 8.24; 95% confidence interval, 2.97–22.89; p<0.001).[129] A higher abundance of *Candida* species enriched at tumor sites is associated with decreased survival across multiple gastrointestinal cancer types, including EAC and ESCC.[22] *Candida* species have been shown to produce carcinogenic concentrations of acetaldehyde following ethanol exposure.[82,130] Notably, *Candida* species have also been shown to produce acetaldehyde when exposed to glucose, revealing an important functional distinction from oropharyngeal bacteria, in which production appears to be dependent on ethanol exposure.[84] This represents another potential mechanism for carcinogenesis via disruption of host metabolic pathways and local tissue injury, although studies have primarily focused on oral cancers, and data in EC are limited.[131,132] Smoking and alcohol consumption may upregulate yeast acetaldehyde production, underscoring the complex interplay between the mycobiome, environmental exposures and host behavior.[131] Although these findings are preliminary, they highlight a potentially important role for fungal-mediated regulation of host immunity and metabolism and support further investigation into the mycobiome as a modulator of immunotherapeutic response in EC.

Future directions

Fecal microbiota transplant (FMT) is under study as a potential intervention to address cancer-related dysbiosis in multiple tumor types.[133] FMT is approved for recurrent *Clostridioides difficile* infection.[134,135] Investigation of FMT in an oncologic context is in its early stages. Two phase I studies conducted by our collaborators, Drs Boursi and Davar, showed that FMT can overcome primary resistance in advanced melanoma. In the first phase I study of 10 patients, FMT restored anti-PD-1 response (partial response n=2, complete response n=1) and increased CD8+ T-cell activation.[136] The second trial demonstrated benefit in 6 of 15 patients with melanoma who received FMT. Responders had enrichment of taxa associated with anti-PD-1 response and changes in proteomic and metabolomic signatures, suggesting beneficial alterations in the tumor microenvironment that promoted anti-PD-1 sensitivity.[137] Subsequent trials in melanoma found positive response rates in treatment-naive patients receiving concurrent donor FMT and anti-PD-1 therapy (objective response rate [ORR], 65%) and dual anti-PD-1 and cytotoxic T-lymphocyte-associated protein 4 (CTLA-4) therapy (ORR, 75% with elimination of deleterious bacterial strains).[138,139] Promising results, albeit in relatively small sample sizes, were seen in other solid tumors, including non-small cell lung cancer and renal cell carcinoma.[139–142] Within the scope of advanced gastrointestinal cancers, two phase I trials have examined FMT in patients resistant to anti-PD-1 therapy, with disease control rates of 46.2% and 40%.[143,144] To date, there are no large-scale randomized controlled trials (RCTs) that investigate FMT in any tumor type, and the trials that have been published largely lack statistical power for assessment of efficacy, creating a barrier to adoption in clinical practice. These phase I and phase II trials have demonstrated the safety and feasibility of FMT, supporting the need for larger RCTs to fully assess the therapeutic potential of FMT.[145] Exactly which microbial populations constitute the optimal FMT donor profile remains to be determined. An additional consideration is the influence of geography, ethnicity, diet and environmental exposures on host microbial populations. These crucial variations may influence both the composition and efficacy of microbiome-based interventions, including FMT, and should be individualized to the patient.

Given the strong evidence for the pro-oncogenic role played by certain bacterial taxa, such as *F. nucleatum,* there may also be therapeutic benefit in the eradication of deleterious bacteria. A meta-analysis of 41,663 patients from 123 cohorts found that administration of antibiotics around the time of ICI treatment was associated with shorter overall survival (hazard ratio [HR], 1.61; 95% CI, 1.48–1.76) and PFS (HR, 1.45; 95% CI, 1.32–1.60), suggesting that a broad-spectrum antimicrobial approach would not be optimal.[146] Using antibiotics that specifically target deleterious bacteria may represent an alternative approach. In a retrospective analysis of 36,105 patients with colorectal cancer, those who received antibiotic therapy targeting anaerobic bacteria had a 25.5% lower rate of cancer recurrence or death (HR, 0.745; 95% CI, 0.57–0.98).[147] Targeted drug delivery may be a solution to the off-target effects of broad-spectrum antibiotics, sparing host microbial populations and preserving immune function. PEGylated metronidazole, a nano-antibiotic designed to target intratumoral *F. nucleatum* while leaving the gastrointestinal microbiome undisturbed, augmented the effects of oxaliplatin and immunotherapy on colorectal cancer cells *in vitro,* suggesting a reversal of bacteria-induced drug resistance.[148] Similarly, the use of a silver–tinidazole liposomal complex targeted to intratumoral bacteria resulted in greater duration of survival in colorectal cancer murine models colonized with *F. nucleatum*. Antibiotic treatment led to the generation of microbial neoantigens, priming CD8+ T cells to recognize and target both infected and uninfected tumor tissue.[147] An alternative strategy for targeted antimicrobial activity may be found in bacteriophages, which are viruses that infect and eliminate specific bacteria. In colorectal cancer models, a bacteriophage successfully targeted *F. nucleatum*, resulting in activation of the host immune response against tumor cells *in vitro* and prolonged survival in murine models.[149] Although the role of targeted antibacterial therapy has only been explored in the preclinical settings, further investigation is warranted, particularly given the beneficial activation of host antitumor immune response observed in multiple studies after *F. nucleatum* elimination.

One potential application of this literature is biomarker development. EAC and ESCC are frequently diagnosed at advanced clinical stages after years of silent progression, generating strong interest in non-invasive or minimally invasive sampling that could improve detection or risk stratification. Several studies have shown distinct microbial signatures in oral samples in ESCC and EAC alike, with advanced neoplasia characterized by greater dysbiosis and microbial metabolic shifts.[6,8,150] Similarly, fecal sampling in ESCC has identified certain patterns of microbial enrichment that appear consistently across multiple studies, as well as serum antigen testing in the case of *F. nucleatum*.[32–34] Although further investigation is necessary before clinical implementation, oral and fecal sampling offer important practical advantages over invasive endoscopic biopsy, particularly by enabling less invasive longitudinal monitoring and potentially more feasible large-scale screening strategies. Direct esophageal cell sampling via non-endoscopic 'swallowed capsule sponge' devices could also represent a less invasive method for bacterial and fungal sampling, although this approach has not been directly investigated.[151] For EAC specifically, the most realistic near-term use of the microbiome is as one component of a multimodal risk framework rather than a stand-alone test. Future models will likely need to integrate

microbial signatures with established clinical and pathologic variables, such as reflux burden, obesity, BE segment length, dysplasia grade and known molecular abnormalities. This is especially true because the EAC microbiome literature has not yet produced a universally reproducible single-organism marker, although it repeatedly demonstrates ecosystem-level disturbances across the progression sequence.[24] In contrast, several compelling bacterial biomarker candidates have emerged in ESCC, particularly *F. nucleatum*, supporting the rationale for larger prospective studies to validate their clinical utility and determine their potential roles in detection, treatment prediction and disease monitoring.

Conclusion

Esophageal cancers (EAC and ESCC) are prevalent worldwide and are associated with a poor prognosis despite recent advances, including the use of ICIs. Dysbiosis is associated with carcinogenesis, tumor progression and treatment response. Alterations in microbial populations and taxonomic diversity appear in both disease subtypes. *Fusobacterium* is a prominent bacterium in ESCC, with implications for tumorigenesis, inflammatory signaling in the tumor microenvironment, immune evasion, treatment resistance and poor clinical outcomes. The fungal mycobiome represents a novel and underexplored dimension of EC biology, highlighting the complexity of bacteria–fungi–host interactions. Further investigation of microbiome profiles and microbial biomarker candidates in EAC and ESCC is crucial, given their potential implications for early detection and the prediction of therapeutic response. Emerging microbiome-directed interventions, including FMT and targeted antimicrobial elimination, may represent potential future therapeutic targets for EC. ❑

1. Sung H, Filho AM, Laversanne M, et al. Global cancer statistics 2024: GLOBOCAN estimates of incidence and mortality worldwide for 34 cancers in 186 countries. *CA Cancer J Clin*. 2026;76:e70090. DOI: 10.3322/caac.70090.
2. Morgan E, Soerjomataram I, Rumgay H, et al. The global landscape of esophageal squamous cell carcinoma and esophageal adenocarcinoma incidence and mortality in 2020 and projections to 2040: New estimates from GLOBOCAN 2020. *Gastroenterology*. 2022;163:649–58.. DOI: 10.1053/j.gastro.2022.05.054.
3. NIH National Cancer Institute Surveillance, Epidemiology and End Results Program. Cancer of the Esophagus – Cancer Stat Facts Available at: https://seer.cancer.gov/statfacts/html/esoph.html (accessed: 12 May 2026).
4. Abnet CC, Arnold M, Wei W-Q. Epidemiology of esophageal squamous cell carcinoma. *Gastroenterology*. 2018;154:360–73. DOI: 10.1053/j.gastro.2017.08.023.
5. Uhlenhopp DJ, Then EO, Sunkara T, Gaduputi V. Epidemiology of esophageal cancer: Update in global trends, etiology and risk factors. *Clin J Gastroenterol*. 2020;13:1010–21. DOI: 10.1007/s12328-020-01237-x.
6. Snider EJ, Compres G, Freedberg DE, et al. Alterations to the esophageal microbiome associated with progression from Barrett's esophagus to esophageal adenocarcinoma. *Cancer Epidemiol Biomarkers Prev*. 2019;28:1687–93. DOI: 10.1158/1055-9965.EPI-19-0008.
7. Li Y, Xing S, Chen F, et al. Intracellular *Fusobacterium nucleatum* infection attenuates antitumor immunity in esophageal squamous cell carcinoma. *Nat Commun*. 2023;14:5788. DOI: 10.1038/s41467-023-40987-3.
8. Ohsawa M, Nishi H, Hamai Y, et al. Relationship between the oral microbiome and treatment efficacy in esophageal squamous cell carcinoma. *Ann Surg Oncol*. 2026;33:3203–13. DOI: 10.1245/s10434-025-18945-8.
9. Wu H, Liu Q, Li J, et al. Tumor-resident microbiota-based risk model predicts neoadjuvant therapy response of locally advanced esophageal squamous cell carcinoma patients. *Adv Sci*. 2024;11:2309742. DOI: 10.1002/advs.202309742.
10. Li N, Gao L, Ge Y, et al. Impact of the gut microbiome on response and toxicity to chemotherapy in advanced esophageal cancer. *Heliyon*. 2024;10:e32770. DOI: 10.1016/j.heliyon.2024.e32770.
11. Yamamura K, Izumi D, Kandimalla R, et al. Intratumoral *Fusobacterium nucleatum* levels predict therapeutic response to neoadjuvant chemotherapy in esophageal squamous cell carcinoma. *Clin Cancer Res*. 2019;25:6170–9. DOI: 10.1158/1078-0432.CCR-19-0318.
12. Liu Y, Baba Y, Ishimoto T, et al. *Fusobacterium nucleatum* confers chemoresistance by modulating autophagy in oesophageal squamous cell carcinoma. *Br J Cancer*. 2021;124:963–74. DOI: 10.1038/s41416-020-01198-5.
13. Yoshikawa T, Nishi H, Emi M, et al. Serum IgG titer findings for *Fusobacterium nucleatum* associated with clinical outcome following surgery in patients with esophageal squamous cell carcinoma. *PLoS One*. 2025;20:e0336219. DOI: 10.1371/journal.pone.0336219.
14. Xu L, Qi Y, Jiang Y, et al. Crosstalk between the gut microbiome and clinical response in locally advanced thoracic esophageal squamous cell carcinoma during neoadjuvant camrelizumab and chemotherapy. *Ann Transl Med*. 2022;10:325–325. DOI: 10.21037/atm-22-1165.
15. Zhou J, Zeng X, Sun J, et al. Gut microbiota-derived indole-3-lactic acid suppresses anti-PD-1 efficacy in esophageal squamous cell carcinoma. *Cell Host Microbe*. 2026;34:639–56. DOI: 10.1016/j.chom.2026.02.019.
16. Wu H, Leng X, Liu Q, et al. Intratumoral microbiota composition regulates chemoimmunotherapy response in esophageal squamous cell carcinoma. *Cancer Res*. 2023;83:3131–44. DOI: 10.1158/0008-5472.CAN-22-2593.
17. van den Ende T, de Clercq NC, Davids M, et al. Fecal, duodenal, and tumor microbiota composition of esophageal carcinoma patients, a longitudinal prospective cohort. *J Natl Cancer Inst*. 2024;116:1834–44. DOI: 10.1093/jnci/djae153.
18. Shaikh FY, Lee S, White JR, et al. Fecal microbiome composition correlates with pathologic complete response in patients with operable esophageal cancer treated with combined chemoradiotherapy and immunotherapy. *Cancers (Basel)*. 2024;16:3644. DOI: 10.3390/cancers16213644.
19. Yuan L, Zhang Y, Wen C, et al. Esophageal cancer and precancerous lesions: Focus on resident bacteria and fungi. *Microbiol Spectr*. 2025;13:e0313724. DOI: 10.1128/spectrum.03137-24.
20. Rao W-Q, Lin Z, Jiang J, et al. Esophageal mycobiome landscape and interkingdom interactions in esophageal squamous cell carcinoma. *Gastroenterol Rep (Oxf)*. 2023;11:goad022. DOI: 10.1093/gastro/goad022.
21. Liang L, Mai S, Sethi G, et al. Fungal microbiota signatures anticipate neoadjuvant immunochemotherapy outcomes in esophageal cancer. *J Immunother Cancer*. 2025;13:e011508. DOI: 10.1136/jitc-2025-011508.
22. Dohlman AB, Klug J, Mesko M, et al. A pan-cancer mycobiome analysis reveals fungal involvement in gastrointestinal and lung tumors. *Cell*. 2022;185:3807–22.. DOI: 10.1016/j.cell.2022.09.015.
23. Yang L, Lu X, Nossa CW, et al. Inflammation and intestinal metaplasia of the distal esophagus are associated with alterations in the microbiome. *Gastroenterology*. 2009;137:588–97. DOI: 10.1053/j.gastro.2009.04.046.
24. Lopetuso LR, Severgnini M, Pecere S, et al. Esophageal microbiome signature in patients with Barrett's esophagus and esophageal adenocarcinoma. *PLoS One*. 2020;15:e0231789. DOI: 10.1371/journal.pone.0231789.
25. Hao Y, Karaoz U, Yang L, et al. Progressive dysbiosis of human orodigestive microbiota along the sequence of gastroesophageal reflux, Barrett's esophagus and esophageal adenocarcinoma. *Int J Cancer*. 2022;151:1703–16. DOI: 10.1002/ijc.34191.
26. Elliott DRF, Walker AW, O'Donovan M, et al. A non-endoscopic device to sample the oesophageal microbiota: A case-control study. *Lancet Gastroenterol Hepatol*. 2017;2:32–42. DOI: 10.1016/S2468-1253(16)30086-3.
27. Deshpande NP, Riordan SM, Gorman CJ, et al. Multi-omics of the esophageal microenvironment identifies signatures associated with progression of Barrett's esophagus. *Genome Med*. 2021;13:133. DOI: 10.1186/s13073-021-00951-6.
28. Verbeek RE, Siersema PD, Ten Kate FJ, et al. Toll-like receptor 4 activation in Barrett's esophagus results in a strong increase in COX-2 expression. *J Gastroenterol*. 2014;49:1121–34. DOI: 10.1007/s00535-013-0862-6.
29. Mozaffari Namin B, Soltan Dallal MM, Ebrahimi Daryani N. The effect of *Campylobacter concisus* on expression of IL-18, TNF-α and P53 in Barrett's cell lines. *Jundishapur J Microbiol*. 2015;8:e26393. DOI: 10.5812/jjm.26393.
30. Tanes C, Li Y, Falk GW, et al. Increased reflux secondary bile acids are associated with changes to the microbiome and transcriptome in Barrett's esophagus. *Gut Microbes*. 2025;17:2545420. DOI: 10.1080/19490976.2025.2545420.
31. Guccione C, Yadlapati R, Shah S, et al. Challenges in determining the role of microbiome evolution in Barrett's esophagus and progression to esophageal adenocarcinoma. *Microorganisms*. 2021;9:2003. DOI: 10.3390/microorganisms9102003.
32. Marasco G, Colecchia L, Salvi D, et al. The role of microbiota in upper gastrointestinal cancers. *Cancers (Basel)*. 2025;17:1719. DOI: 10.3390/cancers17101719.
33. Liu L, Liang L, Luo Y, et al. Unveiling the power of gut microbiome in predicting neoadjuvant immunochemotherapy responses in esophageal squamous cell carcinoma. *Research (Wash D C)*. 2024;7:0529. DOI: 10.34133/research.0529.
34. Huang X, Chen X, Wan G, et al. Mechanism of intestinal microbiota disturbance promoting the occurrence and development of esophageal squamous cell carcinoma——based on microbiomics and metabolomics. *BMC Cancer*. 2024;24:245. DOI: 10.1186/s12885-024-11982-8.
35. Hasuda H, Ikeda T, Makizaki Y, et al. Alterations in the gut microbiome in patients with esophageal carcinoma in response to esophagectomy and neoadjuvant treatment. *Surg Today*. 2023;53:663–74. DOI: 10.1007/s00595-022-02607-3.
36. Cheung MK, Yue GGL, Lauw S, et al. Alterations in gut microbiota of esophageal squamous cell carcinoma patients. *J Gastroenterol Hepatol*. 2022;37:1919–27. DOI: 10.1111/jgh.15941.
37. Deng Y, Tang D, Hou P, et al. Dysbiosis of gut microbiota in patients with esophageal cancer. *Microb Pathog*. 2021;150:104709. DOI: 10.1016/j.micpath.2020.104709.
38. Gao M, Wu J, Zhou S, et al. Combining fecal microbiome and metabolomics reveals diagnostic biomarkers for esophageal squamous cell carcinoma. *Microbiol Spectr*. 2024;12:e0401223. DOI: 10.1128/spectrum.04012-23.
39. Wu Z, Guo J, Zhang Z, et al. Bacteroidetes promotes esophageal squamous carcinoma invasion and metastasis through LPS-mediated TLR4/Myd88/NF-κB pathway and inflammatory changes. *Sci Rep*. 2024;14:12827. DOI: 10.1038/s41598-024-63774-6.
40. Ishaq HM, Mohammad IS, Sher Muhammad K, et al. Gut microbial dysbiosis and its association with esophageal cancer. *J Appl Biomed*. 2021;19:1–13. DOI: 10.32725/jab.2021.005.
41. Deng J, Huang Y, Yu K, et al. Changes in the gut microbiome of patients with esophageal cancer: A systematic review and meta-analysis based on 16S gene sequencing technology. *Microb Pathog*. 2024;193:106784. DOI: 10.1016/j.micpath.2024.106784.
42. Neurath MF, Artis D, Becker C. The intestinal barrier: A pivotal role in health, inflammation, and cancer. *Lancet Gastroenterol Hepatol*. 2025;10:573–92. DOI: 10.1016/S2468-1253(24)00390-X.
43. Yang H, Cai J, Huang X, et al. Gram-negative microflora dysbiosis facilitates tumor progression and immune evasion by activating the CCL3/CCL5-CCR1-MAPK-PD-L1 pathway in esophageal squamous cell carcinoma. *Mol Cancer Res*. 2025;23:71–85. DOI: 10.1158/1541-7786.MCR-24-0451.
44. Peng Z, Wan P, Deng Y, et al. Lipopolysaccharide exacerbates to the migration, invasion, and epithelial-mesenchymal transition of esophageal cancer cells by TLR4/NF-κb axis. *Environ Toxicol*. 2023;38:1090–9. DOI: 10.1002/tox.23750.
45. Zu Y, Ping W, Deng T, et al. Lipopolysaccharide-induced toll-like receptor 4 signaling in esophageal squamous cell carcinoma promotes tumor proliferation and regulates inflammatory cytokines expression. *Dis Esophagus*. 2017;30:1–8. DOI: 10.1111/dote.12466.
46. Signat B, Roques C, Poulet P, Duffaut D. *Fusobacterium nucleatum* in periodontal health and disease. *Curr Issues Mol Biol*. 2011;13:25–36.
47. Liu Y, Baba Y, Ishimoto T, et al. Progress in characterizing the linkage between *Fusobacterium nucleatum* and gastrointestinal cancer. *J Gastroenterol*. 2019;54:33–41. DOI: 10.1007/s00535-018-1512-9.
48. Nomburg J, Bullman S, Nasrollahzadeh D, et al. An international report on bacterial communities in esophageal squamous cell carcinoma. *Int J Cancer*. 2022;151:1947–59. DOI: 10.1002/ijc.34212.
49. Li M, Shao D, Fan Z, et al. Non-invasive early detection on esophageal squamous cell carcinoma and precancerous lesions by microbial biomarkers combining epidemiological factors in China. *J Gastroenterol*. 2024;59:531–42. DOI: 10.1007/s00535-024-02117-8.
50. Zhang B, Xiao Q, Chen H, et al. Comparison of tumor-associated and nontumor-associated esophageal mucosa microbiota in patients with esophageal squamous cell carcinoma. *Medicine (Baltimore)*. 2022;101:e30483. DOI: 10.1097/MD.0000000000030483.
51. Yang W, Chen C-H, Jia M, et al. Tumor-associated microbiota in esophageal squamous cell carcinoma. *Front Cell Dev Biol*. 2021;9:641270. DOI: 10.3389/fcell.2021.641270.
52. Yamamura K, Baba Y, Nakagawa S, et al. Human microbiome *Fusobacterium nucleatum* in esophageal cancer tissue is associated with prognosis. *Clin Cancer Res*. 2016;22:5574–81. DOI: 10.1158/1078-0432.CCR-16-1786.
53. Shao D, Vogtmann E, Liu A, et al. Microbial characterization of esophageal squamous cell carcinoma and gastric cardia adenocarcinoma from a high-risk region of China. *Cancer*. 2019;125:3993–4002. DOI: 10.1002/cncr.32403.
54. Li Z, Shi C, Zheng J, et al. *Fusobacterium nucleatum* predicts a high risk of metastasis for esophageal squamous cell

carcinoma. *BMC Microbiol*. 2021;21:301. DOI: 10.1186/s12866-021-02352-6.
55. Lei J, Xu F, Deng C, et al. *Fusobacterium nucleatum* promotes the early occurrence of esophageal cancer through upregulation of IL-32/PRTN3 expression. *Cancer Sci*. 2023;114:2414–28. DOI: 10.1111/cas.15787.
56. Ding N, Cheng Y, Liu H, et al. *Fusobacterium nucleatum* infection induces malignant proliferation of esophageal squamous cell carcinoma cell by putrescine production. *Microbiol Spectr*. 2023;11:e0275922. DOI: 10.1128/spectrum.02759-22.
57. Guo S, Chen F, Li L, et al. Intracellular *Fusobacterium nucleatum* infection increases METTL3-mediated m6a methylation to promote the metastasis of esophageal squamous cell carcinoma. *J Adv Res*. 2024;61:165–78. DOI: 10.1016/j.jare.2023.08.014.
58. Nomoto D, Baba Y, Liu Y, et al. *Fusobacterium nucleatum* promotes esophageal squamous cell carcinoma progression via the NOD1/RIPK2/NF-κB pathway. *Cancer Lett*. 2022;530:59–67. DOI: 10.1016/j.canlet.2022.01.014.
59. Mao H, Zhao X, Sun S-C. NF-κb in inflammation and cancer. *Cell Mol Immunol*. 2025;22:811–39. DOI: 10.1038/s41423-025-01310-w.
60. Yin H, Zhang J, Zhang H, et al. *Fusobacterium nucleatum* promotes proliferation in oesophageal squamous cell carcinoma via AHR/CYP1A1 signalling. *FEBS J*. 2023;290:837–54. DOI: 10.1111/febs.16619.
61. How KY, Song KP, Chan KG. *Porphyromonas gingivalis*: An overview of periodontopathic pathogen below the gum line. *Front Microbiol*. 2016;7:53. DOI: 10.3389/fmicb.2016.00053.
62. Gao S, Li S, Ma Z, et al. Presence of *Porphyromonas gingivalis* in esophagus and its association with the clinicopathological characteristics and survival in patients with esophageal cancer. *Infect Agent Cancer*. 2016;11:3. DOI: 10.1186/s13027-016-0049-x.
63. Li M, Shao D, Zhou J, et al. Signatures within esophageal microbiota with progression of esophageal squamous cell carcinoma. *Chin J Cancer Res*. 2020;32:755–67. DOI: 10.21147/j.issn.1000-9604.2020.06.09.
64. Chen M-F, Lu M-S, Hsieh C-C, Chen W-C. *Porphyromonas gingivalis* promotes tumor progression in esophageal squamous cell carcinoma. *Cell Oncol (Dordr)*. 2021;44:373–84. DOI: 10.1007/s13402-020-00573-x.
65. Gao S, Liu K, Jiao Y, et al. Selective activation of TGFβ signaling by *P. gingivalis*-mediated upregulation of GARP aggravates esophageal squamous cell carcinoma. *Am J Cancer Res*. 2023;13:2013–29.
66. Yilmaz O, Jungas T, Verbeke P, Ojcius DM. Activation of the phosphatidylinositol 3-kinase/akt pathway contributes to survival of primary epithelial cells infected with the periodontal pathogen *Porphyromonas gingivalis*. *Infect Immun*. 2004;72:3743–51. DOI: 10.1128/IAI.72.7.3743-3751.2004.
67. Yao L, Jermanus C, Barbetta B, et al. *Porphyromonas gingivalis* infection sequesters pro-apoptotic bad through akt in primary gingival epithelial cells. *Mol Oral Microbiol*. 2010;25:89–101. DOI: 10.1111/j.2041-1014.2010.00569.x.
68. Morandini AC, Ramos-Junior ES, Potempa J, et al. *Porphyromonas gingivalis* fimbriae dampen P2X7-dependent interleukin-1β secretion. *J Innate Immun*. 2014;6:831–45. DOI: 10.1159/000363338.
69. Yilmaz O, Yao L, Maeda K, et al. ATP scavenging by the intracellular pathogen *Porphyromonas gingivalis* inhibits P2X7-mediated host-cell apoptosis. *Cell Microbiol*. 2008;10:863–75. DOI: 10.1111/j.1462-5822.2007.01089.x.
70. Gao S, Liu Y, Duan X, et al. *Porphyromonas gingivalis* infection exacerbates oesophageal cancer and promotes resistance to neoadjuvant chemotherapy. *Br J Cancer*. 2021;125:433–44. DOI: 10.1038/s41416-021-01419-5.
71. Lu C, Chen Z, Lu H, Zhao K. *Porphyromonas gingivalis* lipopolysaccharide regulates cell proliferation, apoptosis, autophagy in esophageal squamous cell carcinoma via TLR4/MYD88/JNK pathway. *J Clin Biochem Nutr*. 2024;74:213–20. DOI: 10.3164/jcbn.22-138.
72. Yang R, Gu B, Jiang Q, et al. *Porphyromonas gingivalis*-induced HuR upregulation suppresses SLC39A10-derived hsa_circ_0057552 in esophageal squamous cell carcinoma. *Microbes Infect*. 2025;27:105548. DOI: 10.1016/j.micinf.2025.105548.
73. Hong K, Zhang H, Qi J, et al. *Porphyromonas gingivalis* extracellular vesicles promotes tumor metastasis in esophageal squamous cell carcinoma by inducing PRKACB/JNK/ NFATC2 axis. *J Nanobiotechnol*. 2025;24:66. DOI: 10.1186/s12951-025-03932-8.
74. Qi Y-J, Jiao Y-L, Chen P, et al. Autophagy induced by *Porphyromonas gingivalis* aggravates esophageal squamous cell carcinoma via YAP/TAZ-mir-21-5p axis. *Cell Signal*. 2025;135:111974. DOI: 10.1016/j.cellsig.2025.111974.
75. Sun M, Peng Z, Shen W, et al. Synergism of *Fusobacterium periodonticum* and N-nitrosamines promote the formation of EMT subtypes in ESCC by modulating wnt3a palmitoylation. *Gut Microbes*. 2024;16:2391521. DOI: 10.1080/19490976.2024.2391521.
76. Guo X, Wan P, Shen W, et al. *Fusobacterium periodonticum* BCT protein targeting glucose metabolism to promote the epithelial-mesenchymal transition of esophageal cancer cells by lactic acid. *J Transl Med*. 2024;22:401. DOI: 10.1186/s12967-024-05157-z.
77. Lu R, Zhu J, Song Q, et al. Targeting intratumoral *Streptococcus mitis* suppresses the progression of esophageal squamous cell carcinoma. *Sci China Life Sci*. 2026;69:1238–54. DOI: 10.1007/s11427-025-3097-2.
78. Narikiyo M, Tanabe C, Yamada Y, et al. Frequent and preferential infection of *Treponema denticola*, *Streptococcus mitis*, and *Streptococcus anginosus* in esophageal cancers. *Cancer Sci*. 2004;95:569–74. DOI: 10.1111/j.1349-7006.2004.tb02488.x.
79. Liu Y, Lin Z, Lin Y, et al. *Streptococcus* and *Prevotella* are associated with the prognosis of oesophageal squamous cell carcinoma. *J Med Microbiol*. 2018;67:1058–68. DOI: 10.1099/jmm.0.000754.
80. Greathouse KL, Stone JK, Vargas AJ, et al. Co-enrichment of cancer-associated bacterial taxa is correlated with immune cell infiltrates in esophageal tumor tissue. *Sci Rep*. 2024;14:2574. DOI: 10.1038/s41598-023-48862-3.
81. Amanuma Y, Ohashi S, Itatani Y, et al. Protective role of ALDH2 against acetaldehyde-derived DNA damage in oesophageal squamous epithelium. *Sci Rep*. 2015;5:14142. DOI: 10.1038/srep14142.
82. Nieminen MT, Salaspuro M. Local acetaldehyde-an essential role in alcohol-related upper gastrointestinal tract carcinogenesis. *Cancers (Basel)*. 2018;10:11. DOI: 10.3390/cancers10010011.
83. Zhu H, Yip HC, Cheung MK, et al. Convergent dysbiosis of upper aerodigestive microbiota between patients with esophageal and oral cavity squamous cell carcinoma. *Int J Cancer*. 2023;152:1903–15. DOI: 10.1002/ijc.34460.
84. Moritani K, Takeshita T, Shibata Y, et al. Acetaldehyde production by major oral microbes. *Oral Dis*. 2015;21:748–54. DOI: 10.1111/odi.12341.
85. Flashner S, Shimonosono M, Tomita Y, et al. ALDH2 dysfunction and alcohol cooperate in cancer stem cell enrichment. *Carcinogenesis*. 2024;45:95–106. DOI: 10.1093/carcin/bgad085.
86. Yao C-C, Tai W-C, Liang C-M, et al. Alternation of the oral, gastric and stool microbiome in patients with reflux esophagitis and Barrett's esophagus. *J Microbiol Immunol Infect*. 2026;59:463–70. DOI: 10.1016/j.jmii.2025.12.007.
87. Kilian M, Chapple ILC, Hannig M, et al. The oral microbiome–an update for oral healthcare professionals. *Br Dent J*. 2016;221:657–66. DOI: 10.1038/sj.bdj.2016.865.
88. Invernici MM, Salvador SL, Silva PHF, et al. Effects of *Bifidobacterium* probiotic on the treatment of chronic periodontitis: A randomized clinical trial. *J Clinic Periodontology*. 2018;45:1198–210. DOI: 10.1111/jcpe.12995.
89. Jackson DN, Theiss AL. Gut bacteria signaling to mitochondria in intestinal inflammation and cancer. *Gut Microbes*. 2020;11:285–304. DOI: 10.1080/19490976.2019.1592421.
90. Liu Y, Zhou F, Yang H, et al. *Porphyromonas gingivalis* promotes malignancy and chemo-resistance via GSK3β-mediated mitochondrial oxidative phosphorylation in human esophageal squamous cell carcinoma. *Transl Oncol*. 2023;32:101656. DOI: 10.1016/j.tranon.2023.101656.
91. Ofuchi T, Hu Q, Omachi K, et al. Intratumoral *Fusobacterium nucleatum* drives cancer-associated fibroblasts enrichment and immune exclusion in esophageal squamous cell carcinoma. *Ann Gastroenterol Surg*. 2026;10:611–20. DOI: 10.1002/ags3.70116.
92. Li Y, Yang HS, Klasse PJ, Zhao Z. The significance of antigen-antibody-binding avidity in clinical diagnosis. *Crit Rev Clin Lab Sci*. 2025;62:9–23. DOI: 10.1080/10408363.2024.2379286.
93. Yu T, Guo F, Yu Y, et al. *Fusobacterium nucleatum* promotes chemoresistance to colorectal cancer by modulating autophagy. *Cell*. 2017;170:548–563.. DOI: 10.1016/j.cell.2017.07.008.
94. Geller LT, Barzily-Rokni M, Danino T, et al. Potential role of intratumor bacteria in mediating tumor resistance to the chemotherapeutic drug gemcitabine. *Science*. 2017;357:1156–60. DOI: 10.1126/science.aah5043.
95. Liang M, Liu Y, Zhang Z, et al. *Fusobacterium nucleatum* induces MDSCs enrichment *via* activation the NLRP3 inflammosome in ESCC cells, leading to cisplatin resistance. *Ann Med*. 2022;54:989–1003. DOI: 10.1080/07853890.2022.2061045.
96. Zhang J-W, Zhang D, Yin H-S, 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;15:2197836. DOI: 10.1080/19490976.2023.2197836.
97. Shah MA, Kennedy EB, Alarcon-Rozas AE, et al. Immunotherapy and targeted therapy for advanced gastroesophageal cancer: ASCO guideline. *J Clin Oncol*. 2023;41:1470–91. DOI: 10.1200/JCO.22.02331.
98. Sun J-M, Shen L, Shah MA, et al. Pembrolizumab plus chemotherapy versus chemotherapy alone for first-line treatment of advanced oesophageal cancer (KEYNOTE-590): A randomised, placebo-controlled, phase 3 study. *Lancet*. 2021;398:759–71. DOI: 10.1016/S0140-6736(21)01234-4.
99. Doki Y, Ajani JA, Kato K, et al. Nivolumab combination therapy in advanced esophageal squamous-cell carcinoma. *N Engl J Med*. 2022;386:449–62. DOI: 10.1056/NEJMoa2111380.
100. Xu J, Kato K, Raymond E, et al. Tislelizumab plus chemotherapy versus placebo plus chemotherapy as first-line treatment for advanced or metastatic oesophageal squamous cell carcinoma (RATIONALE-306): A global, randomised, placebo-controlled, phase 3 study. *Lancet Oncol*. 2023;24:483–95. DOI: 10.1016/S1470-2045(23)00108-0.
101. Qin J, Xue L, Hao A, et al. Neoadjuvant chemotherapy with or without camrelizumab in resectable esophageal squamous cell carcinoma: The randomized phase 3 ESCORT-NEO/NCCES01 trial. *Nat Med*. 2024;30:2549–57. DOI: 10.1038/s41591-024-03064-w.
102. Weidenbaum C, Gibson MK. Approach to localized squamous cell cancer of the esophagus. *Curr Treat Options Oncol*. 2022;23:1370–87. DOI: 10.1007/s11864-022-01003-w.
103. Papanicolas LE, Sims SK, Taylor SL, et al. Conventional myelosuppressive chemotherapy for non-haematological malignancy disrupts the intestinal microbiome. *BMC Cancer*. 2021;21:591. DOI: 10.1186/s12885-021-08296-4.
104. Bule P, Aguiar SI, Aires-Da-Silva F, Dias JNR. Chemokine-directed tumor microenvironment modulation in cancer immunotherapy. *Int J Mol Sci*. 2021;22:9804. DOI: 10.3390/ijms22189804.
105. Wheless MC, Comer M, Gibson MK. Evolving treatment landscape for advanced esophageal and gastroesophageal junction adenocarcinoma. *Curr Oncol Rep*. 2024;26:1469–88. DOI: 10.1007/s11912-024-01607-5.
106. Nilsson RH, Anslan S, Bahram M, et al. Mycobiome diversity: High-throughput sequencing and identification of fungi. *Nat Rev Microbiol*. 2019;17:95–109. DOI: 10.1038/s41579-018-0116-y.
107. Briard B, Fontaine T, Kanneganti T-D, et al. Fungal cell wall components modulate our immune system. *Cell Surf*. 2021;7:100067. DOI: 10.1016/j.tcsw.2021.100067.
108. Rizzetto L, Ifrim DC, Moretti S, et al. Fungal chitin induces trained immunity in human monocytes during cross-talk of the host with *Saccharomyces cerevisiae*. *J Biol Chem*. 2016;291:7961–72. DOI: 10.1074/jbc.M115.699645.
109. Zhu F, Willette-Brown J, Song N-Y, et al. Autoreactive T cells and chronic fungal infection drive esophageal carcinogenesis. *Cell Host & Microbe*. 2017;21:478–493.. DOI: 10.1016/j.chom.2017.03.006.
110. Liew W-P-P, Mohd-Redzwan S. Mycotoxin: Its impact on gut health and microbiota. *Front Cell Infect Microbiol*. 2018;8:60. DOI: 10.3389/fcimb.2018.00060.
111. Kaźmierczak-Siedlecka K, Dvořák A, Folwarski M, et al. Fungal gut microbiota dysbiosis and its role in colorectal, oral, and pancreatic carcinogenesis. *Cancers (Basel)*. 2020;12:1326. DOI: 10.3390/cancers12051326.
112. Coker OO, Nakatsu G, Dai RZ, et al. Enteric fungal microbiota dysbiosis and ecological alterations in colorectal cancer. *Gut*. 2019;68:654–62. DOI: 10.1136/gutjnl-2018-317178.
113. Aykut B, Pushalkar S, Chen R, et al. The fungal mycobiome promotes pancreatic oncogenesis via activation of MBL. *Nature*. 2019;574:264–7. DOI: 10.1038/s41586-019-1608-2.
114. Saleh I, Zeidan R, Abu-Dieyeh M. The characteristics, occurrence, and toxicological effects of alternariol: A mycotoxin. *Arch Toxicol*. 2024;98:1659–83. DOI: 10.1007/s00204-024-03743-0.
115. Liu GT, Qian YZ, Zhang P, et al. Etiological role of *Alternaria alternata* in human esophageal cancer. *Chin Med J (Engl)*. 1992;105:394–400.
116. Liu GT, Qian YZ, Zhang P, et al. Relationships between *Alternaria alternata* and oesophageal cancer. *IARC Sci Publ*. 1991;258–62.
117. Dong ZG, Liu GT, Dong ZM, et al. Induction of mutagenesis and transformation by the extract of *Alternaria alternata* isolated from grains in Linxian, China. *Carcinogenesis*. 1987;8:989–91. DOI: 10.1093/carcin/8.7.989.
118. Solhaug A, Eriksen GS, Holme JA. Mechanisms of action and toxicity of the mycotoxin alternariol: A review. *Basic Clin Pharmacol Toxicol*. 2016;119:533–9. DOI: 10.1111/bcpt.12635.
119. Liu N-N, Yi C-X, Wei L-Q, et al. The intratumor mycobiome promotes lung cancer progression via myeloid-derived suppressor cells. *Cancer Cell*. 2023;41:1927–1944.. DOI: 10.1016/j.ccell.2023.08.012.
120. Kabwe MH, Vikram S, Mulaudzi K, et al. The gut mycobiota of rural and urban individuals is shaped by geography. *BMC Microbiol*. 2020;20:257. DOI: 10.1186/s12866-020-01907-3.
121. Miceli MH, Díaz JA, Lee SA. Emerging opportunistic yeast infections. *Lancet Infect Dis*. 2011;11:142–51. DOI: 10.1016/S1473-3099(10)70218-8.
122. Liu Y, Koh CMJ, Yap SA, et al. Understanding and exploiting the fatty acid desaturation system in *Rhodotorula toruloides*. *Biotechnol Biofuels*. 2021;14:73. DOI: 10.1186/s13068-021-01924-y.
123. Hsiao HY, Chiang LC, Ueng PP, Tsao GT. Sequential utilization of mixed monosaccharides by yeasts. *Appl Environ Microbiol*. 1982;43:840–5. DOI: 10.1128/aem.43.4.840-845.1982.
124. Rangel-Grimaldo M, Rivero-Cruz I, Madariaga-Mazón A, et al. α-Glucosidase inhibitors from *Preussia minimoides*. *J Nat Prod*. 2017;80:582–7. DOI: 10.1021/acs.jnatprod.6b00574.
125. Rodríguez-Cuesta J, Hernando FL, Mendoza L, et al. *Candida albicans* enhances experimental hepatic melanoma metastasis. *Clin Exp Metastasis*. 2010;27:35–42. DOI: 10.1007/s10585-009-9300-9.
126. Deng W, Su Z, Liang P, et al. Single-cell immune checkpoint landscape of PBMCS stimulated with *Candida albicans*. *Emerg Microbes Infect*. 2021;10:1272–83. DOI: 10.1080/22221751.2021.1942228.
127. Nikou S-A, Zhou C, Griffiths JS, et al. The *Candida albicans* toxin candidalysin mediates distinct epithelial inflammatory responses through P38 and EGFR-ERK pathways. *Sci Signal*. 2022. DOI: 10.1126/scisignal.abj6915.
128. Wurster S, Albert ND, Kontoyiannis DP. *Candida auris* bloodstream infection induces upregulation of the PD-1/PD-L1 immune checkpoint pathway in an immunocompetent mouse model. *mSphere*. 2022;7:e0081721. DOI: 10.1128/msphere.00817-21.
129. Guo X, Lam SY, Janmaat VT, et al. Esophageal candida infection and esophageal cancer risk in patients with Achalasia. *JAMA Netw Open*. 2025;8:e2454685. DOI: 10.1001/jamanetworkopen.2024.54685.
130. Tillonen J, Homann N, Rautio M, et al. Role of yeasts in the salivary acetaldehyde production from ethanol among risk groups for ethanol-associated oral cavity cancer. *Alcohol Clin Exp Res*. 1999;23:1409–15.

131. Gainza-Cirauqui ML, Nieminen MT, Novak Frazer L, et al. Production of carcinogenic acetaldehyde by *Candida albicans* from patients with potentially malignant oral mucosal disorders. *J Oral Pathol Med*. 2013;42:243–9. DOI: 10.1111/j.1600-0714.2012.01203.x.
132. Alnuaimi AD, Ramdzan AN, Wiesenfeld D, et al. Candida virulence and ethanol-derived acetaldehyde production in oral cancer and non-cancer subjects. *Oral Dis*. 2016;22:805–14. DOI: 10.1111/odi.12565.
133. Davar D, Zarour HM, Trinchieri G. Improving immunotherapy in solid tumors using FMT. *Cell*. 2026;189:2214–7. DOI: 10.1016/j.cell.2026.03.021.
134. Feuerstadt P, Louie TJ, Lashner B, et al. SER-109, an oral microbiome therapy for recurrent *Clostridioides difficile* infection. *N Engl J Med*. 2022;386:220–9. DOI: 10.1056/NEJMoa2106516.
135. Khanna S, Assi M, Lee C, et al. Placebo-controlled trial with a Bayesian primary analysis for the prevention of recurrent *Clostridioides difficile* infection. *Drugs*. 2022;82:1527–38. DOI: 10.1007/s40265-022-01797-x.
136. Baruch EN, Youngster I, Ben-Betzalel G, et al. Fecal microbiota transplant promotes response in immunotherapy-refractory melanoma patients. *Science*. 2021;371:602–9. DOI: 10.1126/science.abb5920.
137. Davar D, Dzutsev AK, McCulloch JA, et al. Fecal microbiota transplant overcomes resistance to anti-PD-1 therapy in melanoma patients. *Science*. 2021;371:595–602. DOI: 10.1126/science.abf3363.
138. Routy B, Lenehan JG, Miller WH Jr, et al. Fecal microbiota transplantation plus anti-PD-1 immunotherapy in advanced melanoma: A phase I trial. *Nat Med*. 2023;29:2121–32. DOI: 10.1038/s41591-023-02453-x.
139. Duttagupta S, Messaoudene M, Hunter S, et al. Fecal microbiota transplantation plus immunotherapy in non-small cell lung cancer and melanoma: The phase 2 FMT-LUMINate trial. *Nat Med*. 2026;32:1337–50. DOI: 10.1038/s41591-025-04186-5.
140. Xue Y. Fecal Microbiota Transplantation Combined with Anti-PD-(L)1 Inhibitors as First-Line Maintenance Therapy for Advanced Gastric and Non Small Cell Lung Cancer. on ASCO.org. 2024. Available at: www.asco.org/abstracts-presentations/239018 (accessed: 12 May 2026).
141. Porcari S, Ciccarese C, Heidrich V, et al. Fecal microbiota transplantation plus pembrolizumab and axitinib in metastatic renal cell carcinoma: The randomized phase 2 TACITO trial. *Nat Med*. 2026;32:1316–24. DOI: 10.1038/s41591-025-04189-2.
142. Fernandes R, Jabbarizadeh B, Rajeh A, et al. Fecal microbiota transplantation plus immunotherapy in metastatic renal cell carcinoma: The phase 1 PERFORM trial. *Nat Med*. 2026;32:1325–36. DOI: 10.1038/s41591-025-04183-8.
143. Kim Y, Kim G, Kim S, et al. Fecal microbiota transplantation improves anti-PD-1 inhibitor efficacy in unresectable or metastatic solid cancers refractory to anti-PD-1 inhibitor. *Cell Host Microbe*. 2024;32:1380–93. DOI: 10.1016/j.chom.2024.06.010.
144. Zhang Y, Xu X, Wang S, et al. Fecal microbiota transplantation combined with anti-PD-1 therapy in refractory microsatellite-stable gastric cancer: A phase I feasibility and safety study. *J Immunother Cancer*. 2026;14:e013823. DOI: 10.1136/jitc-2025-013823.
145. Wekking D, Ende T van den, Bijlsma MF, et al. Fecal microbiota transplantation to enhance cancer treatment outcomes across different cancer types: A systematic literature review. *Cancer Treat Rev*. 2025;140:103025. DOI: 10.1016/j.ctrv.2025.103025.
146. Crespin A, Le Bescop C, de Gunzburg J, et al. A systematic review and meta-analysis evaluating the impact of antibiotic use on the clinical outcomes of cancer patients treated with immune checkpoint inhibitors. *Front Oncol*. 2023;13:1075593. DOI: 10.3389/fonc.2023.1075593.
147. Wang M, Rousseau B, Qiu K, et al. Killing tumor-associated bacteria with a liposomal antibiotic generates neoantigens that induce anti-tumor immune responses. *Nat Biotechnol*. 2024;42:1263–74. DOI: 10.1038/s41587-023-01957-8.
148. Wang X, Wang Y, Guo C, et al. Targeted depletion of intratumoral bacteria using pegylated metronidazole to augment cancer treatment efficacy. *Biomaterials*. 2026;327:123772. DOI: 10.1016/j.biomaterials.2025.123772.
149. Dong X, Pan P, Zheng D-W, et al. Bioinorganic hybrid bacteriophage for modulation of intestinal microbiota to remodel tumor-immune microenvironment against colorectal cancer. *Sci Adv*. 2020;6:eaba1590. DOI: 10.1126/sciadv.aba1590.
150. Solfisburg QS, Baldini F, Baldwin-Hunter B, et al. The salivary microbiome and predicted metabolite production are associated with Barrett's esophagus and high-grade dysplasia or adenocarcinoma. *Cancer Epidemiol Biomarkers Prev*. 2024;33:371–80. DOI: 10.1158/1055-9965.EPI-23-0652.
151. Comer M, Chen H, Wheless MC, Gibson MK. Current and novel treatment approaches for locally advanced esophageal squamous cell carcinoma: A narrative review. *Ann Esophagus*. 2025;8:21–21. DOI: 10.21037/aoe-25-12.