

# Microbiology and Lung Cancer: A Comprehensive Review

Xinran Guo<sup>1,\*</sup>, Yue Liu<sup>2</sup>

<sup>1</sup> Tianjin Medical University;

<sup>2</sup> Tianjin Hospital

**Abstract:** There are hundreds of billions of bacterial flora distributed in the human skin, oral cavity, nasal cavity, stomach, small intestine, large intestine, urinary tract and vagina. There are currently more than 1,000 known human microbiota species. The microbiome lives symbiotically with our bodies and is a diverse community that includes bacteria, fungi, and viruses. It plays an important role in maintaining the body's internal balance, adjusting the immune system, and influencing genetic factors. Lung cancer ranks first among new malignant tumors and is also the leading cause of death from malignant tumors. Like most cancers, lung cancer is caused by multiple factors and has a complex pathogenesis, which may include susceptible hosts, environmental factors, chronic disease burden, habits such as smoking and drinking, and other unidentified causes. In recent years, with the development of detection technology and the in-depth understanding of the tumor microenvironment, more and more evidence has confirmed the existence of bacteria within tumors, which has also triggered further in-depth research in this field. Preliminary epidemiological studies have suggested a link between microbes and cancer. This review introduces the sources of microorganisms and their locations, outlines the relationship between lung, oral, and intestinal microbiota and lung cancer and biomolecular mechanisms, and summarizes methods to treat and prevent lung cancer by utilizing these characteristic mechanisms.

## 1. Text

### 1.1 Source of lung microorganisms and location of microorganism sand tumors

In today's society, as people's average life span increases, lifestyle changes and pollutant levels increase, the incidence of cancer continues to increase. There is no doubt that lung cancer is the most threatening among many types of cancer, with its morbidity and mortality ranking first among malignant tumors. According to a data report on the number of new tumors in humanity in 2020 provided by the WHO IARC, the number of new lung cancer cases in the world this year reached approximately 22 million (the second largest is breast malignant tumors), and The death toll exceeds 1.8 million people (this is far more than any other type of cancer); one thing that needs to be pointed out in particular is that most people with lung cancer have smoked; however, there are 100% Non-smokers aged 10 to 15 may also develop this disease. In addition, the incidence rate of non-smoking women is higher than that of men, and the onset of the disease for a shorter time is also a problem that cannot be ignored. There are many reasons for this phenomenon, such as exposure to external harmful substances such as passive smoking, radiation exposure, etc., or air quality problems in living and working places, etc., which can be potential influencing factors - although we are not yet sure. What is the specific pathogenesis of lung cancer in patients who do not smoke. Zhang et al. obtained fresh frozen tumor tissue and matched germline DNA from lung cancer patients who had never received treatment, never smoked, and were exposed to unknown lung cancer risk factors. Through high-coverage whole-genome sequencing of never-smoking lung cancer patients, Zhang et al. 3 subtypes defined by copy number aberrations. Among lung cancer cases from smokers, most subtypes are uncommon and are characterized by UBA1 mutations in somatic cells, germline AR gene changes, and stem cell properties such as lower mutational burden and higher internal diversity. fusion, longer chromosome ends, frequent KRAS mutations, and slow growth are all indicators of possible cancer risk. Other subtypes are characterized by specific amplifications and mutations of EGFR (moderate intensity) and genome-wide doubling (strong). Even in the case of exposure to secondhand smoke, no strong signs of smoking were detected [1]. The major subtype variations in the DNA sequences of non-smoking lung cancer patients are

significantly different from those of smoking lung cancer patients, which opens the way for personalized treatment of non-smoking lung cancer patients. Under normal circumstances, the human body is in a dynamic balance, and microorganisms in various body parts can interact with each other, or indirectly through inflammatory substances, cytokines and metabolites in the systemic circulation. The oral microbiome may be a major source of the lung microbiome. Lung cancer risk can be assessed through the diversity of oral microorganisms. Hosgood et al. used a metagenomic shotgun approach to determine the oral microbial community structure and abundance in prediagnostic mouthwash samples from each case and control group. To assess the association between lung cancer risk and oral microbiome alpha diversity metrics and taxon relative abundance, as well as microbiome beta diversity. After testing, it was found that participants with lower alpha microbial diversity were more likely to develop lung cancer than those with higher alpha microbial diversity, while there was no significant case-control difference in beta microbial diversity. This study shows that lower alpha diversity is associated with a higher risk of lung cancer, and the abundance of certain taxa is associated with altered risk. The risk of lung cancer can be assessed by analyzing the diversity of oral microorganisms [2]. Zhang et al. studied the process by which oral and upper respiratory tract microorganisms shape the lung microbiota and found that the high oral input pneumonia group in the hormone replacement therapy group was worse than the low oral input pneumonia group in terms of pro-inflammatory cytokines (interleukin-1 $\alpha$ , $\beta$ ) levels also increased significantly. This suggests that heterogeneity in this shaping process is associated with lung microbiota variation. The study also observed congruence between oral and lung microbiota, manifested by overall synergy in bacterial interaction networks and niche differences represented by *Prevotella* species. It was found that the microbial content in saliva was significantly positively correlated with interleukin-1, and the characteristics of salivary microflora were determined, which can be used to predict the status of lung microflora [3]. These findings allow us to better understand how oral and upper respiratory tract microbes influence the lung microbiota and raise the possibility of developing techniques to assess and regulate lung health by analyzing oral microbes in mouthwash and saliva. Additionally, the respiratory tract and intestines can communicate with each other through biological processes including microsuction and aspiration. The early formation of the microbiota and immune environment in the human respiratory and gastrointestinal tracts may originate from the skin and the external environment. Despite clear differences in microanatomical features, composition, and population dynamics of intestinal and lung microbiota, these two organs share similar

homeostasis and certain physiological characteristics, such as microbiota maturation processes, mucosal immune systems, coevolution, and immune cell communication and ongoing exposure to the external environment. This complex connection between the gut and lung microbiota is known as the "gut-lung axis." Intestinal probiotics are likely to play a role in inhibiting tumorigenesis by regulating the microbiota through the "gut-lung axis" [4]. Xia et al. used the "gut-lung axis" as the theoretical basis to explore the efficacy of "gut-lung axis" microbiota regulation combined with chemotherapy in patients with advanced non-small cell lung cancer. Combined with traditional platinum-based two-drug chemotherapy, the impact of platinum-based combination chemotherapy on the diversity of intestinal flora, stool before and after chemotherapy, and the status and extent of adverse reactions during chemotherapy in lung cancer patients was studied. Observe the efficacy of new treatments in patients with advanced non-small cell lung cancer [5]. Research shows that intestinal-pulmonary microecological balance may become a new target for lung cancer treatment, providing new treatment strategies and basis for lung cancer patients. Changes in the gut microbiota can affect the lung microbiota and vice versa. COVID-19 is associated with widespread gastrointestinal discomfort. Moon et al. found that compared with healthy controls, the intestinal microbiota of COVID-19 patients had lower bacterial diversity and higher relative abundance of opportunistic pathogens. The long-term presence of viral antigens and disruption of mucosal immunity may increase the risk of intestinal microbiota and inflammation, leading to acute pathological outcomes or acute post-COVID-19 symptoms [6]. From the perspective of genetic

evolution, it can be found that some of the intestinal microorganisms in our bodies originate from our ancestors [7]. With changes in socioeconomics, health conditions, and living habits, the original intestinal microbiota is also constantly changing. In this review, we focus on three potential sources of lung microbes: 1) Adverse environmental factors are common causes of lung microbial changes 2) Oral microbes are the main source of lung microbes 3) Intestinal microbes The connection with lung microbes may be established through the "gut-lung axis". Research shows that microorganisms widely exist in tumor tissues, and the microorganisms found in tumors are mainly bacteria. This provides new ideas for early diagnosis, prognosis, and treatment of tumors. Walker et al. summarized the favorable conditions for bacterial colonization in tumors, including vascular disorder, anoxic and nutrient-rich microenvironment [8]. Xie et al. focused on the sources of tumor microbiota, including damaged mucosal barriers, normal adjacent tissues to tumors, and the circulatory system [9]. A landmark study by Nejman et al. showed that intratumoral bacteria are mainly located within tumor and immune cells [10]. More precise localization revealed that bacteria are in many cases perinuclear and diverse even among different cell types. Confirming the location of microorganisms in cancer can better formulate drug regimens and design treatment measures.

## 2. Lung cancer and microbes

Changing the oral microbial composition may trigger various oral problems, such as tooth decay, periodontal disease, and cancer [11]. In addition, studies have revealed the correlation between oral microecology and many lung diseases, such as chronic obstructive pulmonary disease, bronchial asthma, and lung cancer [12]. By examining the upper respiratory tract microbiota, the oral microorganisms *Veillonella* and *Capnocytophaga* were found to be significantly higher in saliva samples from lung cancer patients [13]. In particular, the combination of 16S rRNA gene sequencing findings and qPCR validation studies revealed significantly higher levels of *Capnocytophaga* and *Veillonella* in saliva from lung cancer patients. Yan et al. studied multiple lung cancer patients by using deep sequencing analysis. Patient

saliva microbiota. *Carbondioxide*, *Selenomonas*, *Veillonella*, and *Neisseria* were found to be significantly altered in the saliva of patients with squamous cell carcinoma and adenocarcinoma compared with controls. This can serve as a potential biomarker for disease detection and classification [14]. For the study of lower respiratory tract microbiota, SH Lee et al. collected bronchoalveolar fluid from multiple patients and analyzed the samples by next-generation sequencing based on 16S rRNA gene sequencing. The relative abundance of Firmicutes and tumor markers in patients with lung cancer is significantly increased in patients with lung cancer, and the genera *Veillonella* and *Megalococcus* are significantly more numerous than in patients with benign diseases [15]. Studies have shown that changes in lung microbial composition in patients with lung cancer are significantly related to changes in oral microbial composition. In recent years, microbiota such as intestinal flora have attracted attention in malignancy. Multiple studies have shown that the intestinal microecology of healthy people and lung cancer patients is significantly different. The intestinal microbiota in the human body is an extremely complex ecosystem, which constitutes the main structure of bacteria throughout the body. These microorganisms are distributed in more than 50 categories, including more than 1,500 different species of bacteria, and there are up to 100 trillion bacteria in the large intestine. There are certain differences in the composition of the microbiota between different individuals, which can be divided into pathogenic bacteria and probiotics. Dysbiosis of the bacterial flora can lead to microbial imbalance in pathological results. Microbial communities are unique to each host and can change rapidly due to various environmental factors including diet [16]. Zheng et al. analyzed the intestinal microbiota composition of early-stage lung cancer patients and healthy individuals through 16S rRNA gene sequencing. Significant differences were found between the two groups Firmicutes, Bacteroidetes, Proteobacteria and Actinobacteria. Among them, Proteobacteria were highly abundant in the cancer group. *Ruminococcus* and *Lacnospiraceae* were highly enriched in the cancer group, while fecal

*Escherichia coli*, *Streptococcus*, *Bifidobacterium*, and *Veillonella* were significantly increased in the healthy group, and *Bacteroidetes* were more abundant in the cancer group. phyla and *Proteobacteria*. In contrast, *Firmicutes* and *Actinobacteria* were mainly significantly reduced in the cancer group [17]. Usually, the pathogenic bacteria in lung cancer patients increase, while the probiotic bacteria decrease relatively. Chen et al. studied the intestinal flora status of lung cancer patients after receiving platinum-based chemotherapy. The RT-PCR method was used to compare the changes in the two dominant bacterial flora before and after chemotherapy. As a result, after receiving platinum drug chemotherapy, the number and diversity of intestinal flora in lung cancer patients decreased, and the composition of the bacterial flora also changed. The numbers of *Lactobacilli* and *Bifidobacteria* were also reduced accordingly [18]. Qiu et al. explored the dynamic changes of intestinal flora in patients with non-small cell lung cancer after concurrent chemoradiotherapy and its predictive role in progression-free survival. The composition and function of the intestinal microbiota were analyzed by 16s rRNA gene sequencing and shotgun metagenomics, and the results showed that the number of *Bacteroidetes* and *Proteobacteria* was increased in patients with non-small cell lung cancer, while the number of *Firmicutes* was decreased [19]. However, excessive reduction or increase in the number of certain bacteria is not necessarily beneficial to the treatment of lung cancer. Hopkins et al. used a combination of proton pump inhibitors and first-line atezolizumab to treat patients with non-small cell lung cancer. They found that the use of proton pump inhibitors reduced the alpha diversity of intestinal

microorganisms and increased the number of *Actinomycetes* and *Micrococcaceae*. , *Enterobacteriaceae*, and *Streptococcaceae* relative abundance. Considering that high alpha diversity and high relative abundance of *Lactococcaceae* and *E. coli* are associated with enhanced antitumor immune activity, this study hypothesizes that the use of proton pump inhibitors may lead to changes in the gut microbiota that impair immune checks Efficacy of point inhibitors [20]. Zheng et al. characterized the gut microbiota composition of early-stage lung cancer patients and healthy individuals through 16s rRNA gene sequencing. found significant changes in the microbial composition of lung cancer patients compared with healthy people. There were some low-abundance phyla in the cancer group, but some higher-abundance phyla, such as *Firmicutes*, *Bacteroidetes*, *Proteobacteria*, and *Actinobacteria*, were also significantly different between the two groups. *Proteobacteria* were high in the cancer group. At the genus level, *Ruminococcus*, an uncharacterized genus of the *Enterobacteriaceae* family, and an uncharacterized genus of the *Lacnopiaceae* family were highly enriched in the cancer group, whereas fecal *E. coli*, *Streptococcus*, and *Diplomyces* *Fibrobacteria* and *Veillonella* were significantly increased in the healthy group, and more abundant species in the cancer group were mainly from the phyla *Bacteroidetes* and *Proteobacteria*. In contrast, species significantly reduced in the cancer group were mainly from *Firmicutes* and *Actinobacteria*. In addition, the reduced proportion of *Firmicutes* and *Bacteroidetes* in the lung cancer group may lead to reduced circulating SCFA, thereby affecting host systemic immunity and inflammation [21]. These data suggest that lung cancer may be associated with an increase in pathogens and a decrease in certain probiotic bacteria. The imbalance of microbial flora, changes in the human microenvironment and metabolism are closely related to the occurrence of tumors. Certain microorganisms increase the risk of lung cancer. Although air enters the human body and reaches the lungs through the respiratory tract and stays here for a while to filter harmful substances and transport oxygen to the blood system for use by various parts of the body (i.e., respiratory function), the traditional concept has always regarded healthy lungs as a Sterile environment; however, in recent years, with the advancement of new detection technologies, we have confirmed that there are a large number of pneumonia pathogens that have not been cultured in the laboratory-this means that pneumonia is not just caused by a specific type of virus or other Infectious diseases caused by organisms are a complex and changeable process involving multiple factors such as host immunity and the impact of foreign antibiotics. Shiels et al. conducted a prospective follow-up of the  $\alpha$ -tocopherol and  $\beta$ -carotene cancer prevention study by studying male smokers. Pulmonary tuberculosis was associated with an

increased risk of lung cancer in male smokers, and *Mycobacterium tuberculosis* in the lungs of lung cancer patients was significantly higher. It is higher than that of healthy people and can be used as a marker for early diagnosis of lung cancer [22]. In addition, the  $\alpha$ -diversity of microbiota in lung tumor tissues of lung cancer patients is significantly reduced. Interleukin-17B plays an important role in studying the regulation of lung microecology in the development of pulmonary fibrosis. The interleukin-17 family is rich in diversity. During the occurrence of pulmonary fibrosis, alveolar macrophages "breed" interleukin-17B. Interleukin-17B can directly act on lung epithelial cells, thereby inducing the expression of downstream genes and promoting The recruitment of neutrophils and the differentiation of Th17 cells play an important role in autoimmune diseases, inflammatory diseases and cancer, and can secrete interleukin-17A, ultimately leading to severe inflammatory damage to the

lungs and the occurrence of pulmonary fibrosis. Both interleukin-17B and interleukin-17A were found to be up-regulated in clinical patient samples. By revealing the role of lung flora in regulating pulmonary mucosa-related diseases, it provides new ideas for related research and clinical treatment of diseases [23]. In summary, the lung, oral, and intestinal microbiota of lung cancer patients significantly affect the host's regulatory functions, and the microbiota often exhibits a loss of diversity, a reduction in total bacterial abundance, and changes in bacterial composition. Compelling evidence reveals that changes in microbial communities can trigger cancer processes to adapt to the host's constantly changing internal and environmental conditions.

### 3. Mechanism

Cancer is generally considered a multifactorial disease, and although the association between carcinogenicity and changes in the lung microbiota has been established, it remains unclear whether the lung microbiota is causally related to the development of lung cancer. To address this issue, it is important to elucidate the role of the microbiota in lung cancer development and elucidate the mechanisms by which the microbiota controls tumor initiation and progression. It can provide a basis for exploring targets for lung cancer treatment. The occurrence of cancer is mainly due to disordered proliferation of normal cells, apoptosis and autophagy disorders, leading to inflammation and DNA damage. The possible mechanisms of microbiota in the pathogenesis of lung cancer can be divided into the following points:

#### 3.1 Microbiome imbalance

When the normal microbial community in the human body is affected by external factors and loses balance, it will lead to disorder of the bacterial ecosystem. This will not only increase the risk of infection, but may also trigger the emergence of a variety of diseases, thus posing a threat to people's health. Hideki et al. used patients with ulcerative colitis as research subjects. Analysis of the microflora and organic acids in the feces of the subjects showed that after supplementing with *Bifidobacterium*, the relative proportions of vulvar *Candida* and butyrate in the *Bacteroidetes* family were reduced. concentrations were significantly reduced. Decreased *Bifidobacterium* concentrations and increased *Bacteroidetes* species are clearly related to the severity of ulcerative colitis [24]. Yang et al. used *Clostridium nucleatum* to infect colorectal cancer cells in mice and found that this method increased the growth rate, invasiveness, and ability of the cancer cells to form xenograft tumors in mice. *Fusobacterium nucleatum* activates Toll-like receptor 4 signaling to MYD88, leading to activation of nuclear factor- $\kappa$ B and increased expression of miR21; this miRNA reduces the level of the RAS GTPase RASA1 [25]. It shows that *Fusobacterium nucleatum* can increase the proliferation and invasion activity of colorectal cancer cells and promote the occurrence of cancer. Julio et al. found that reduced abundance of *Methylobacterium* is more likely to cause tumor occurrence in breast cancer patients [26]. Similar phenomena have been found in other cancers. Xu et al confirmed that *H. pylori* infection generally increases the incidence of pancreatic cancer, but the incidence has not been established [27]. Gautam et al. found that

*Chlamydia trachomatis*, *Chlamydia pneumoniae*, and *Chlamydia psittaci* often infect the human respiratory tract and cause chronic obstructive pulmonary disease, asthma, lung

cancer, etc. [28]. Najafi et al. used 16srRNA gene sequencing technology and found that the lung tumor tissues of lung cancer patients include *Actinobacteria*, *Corynebacteriaceae*, and *Halomonadaceae*, as well as the genera *Corynebacterium*, *Lactobacterium*, and *Halomonas*. The relative abundance of several bacterial taxa is significantly reduced, capturing changes in the lung microbiome in lung cancer will be helpful in the treatment of lung cancer [29]. Wang et al. conducted tests on patients with non-small cell lung cancer and healthy subjects. The study showed that the intestinal flora refers to the microorganisms designated to colonize the ileum, colon, etc., including probiotics such as *Lactobacillus* and *Bifidobacterium* that can synthesize essential vitamins in the body. *Bacilli*. At the same time, these microorganisms will affect the absorption and synthesis of intestinal nutrients, as well as the intake of trace elements. Changes in intestinal flora can affect local intestinal homeostasis and systemic homeostasis [30]. Dysbiosis of commensal microbial communities may alter host susceptibility to carcinogens. At the same time, it has been confirmed that the imbalance of symbiotic microbial communities is related to the occurrence and development of various malignant tumors, such as breast cancer. Xue et al. used enzyme-linked immunosorbent assay, 16srRNA gene sequencing technology, Western blot analysis and other methods to investigate the effect of fucoidan on 7, Effects of intestinal flora and intestinal barrier function on 12-dimethylbenzanthracene-induced breast cancer rats. The results showed that the intestinal wall of the model group was damaged, and the addition of fucoidan could improve the composition of fecal microbiota and repair the intestinal barrier function. This study suggested the use of fucoidan as a gut microbiota modulator to potentially prevent breast cancer [31, 32]. In addition, changes in intestinal flora are also related to the occurrence of nasopharyngeal cancer. Jiang et al. compared the differences in intestinal microbiota composition and biological functions between familial nasopharyngeal carcinoma patients (NPCF), sporadic nasopharyngeal carcinoma patients (NPCS) and healthy controls (NOR), using bioinformatics methods. Gut microbiota DNA sequencing and hematology test results were compared between the two groups. The results showed that compared with the NOR group, the intestinal flora structure of patients in the NPCF group and NPCS group changed significantly. *Clostridium mycoides*, *Citrobacter* species, *Veillonella* species, and *Prevotella* species were significantly increased, while *Akkermansia muciniphila* and *Roseburia* species were significantly decreased. In NPC, *C. coccidioides*, *Veillonella dispersum*, and *Klebsiella* species were significantly increased, and *B. adolescentis* was significantly reduced [33]. This discovery may provide new biomarkers for disease risk prediction and screening of high-risk groups, as well as early non-invasive diagnosis of nasopharyngeal cancer. In non-small cell lung cancer, the gut microbiota may alter the inflammatory and immune status by affecting local substance absorption and metabolism, thereby affecting its progression. Therefore, discovering the characteristics of the intestinal flora of patients with NSCLC may help to discover the flora that promote or inhibit the development of the disease [34]. Additionally, this may help guide the development of probiotic formulations for NSCLC patients.

### 3.2 Immune response

Disturbance of the microbiota can mediate the body's immune response. The occurrence of superficial borderline tumors is often associated with disruption of the host mucosal immune barrier. Rooks et al. used nonradioactive in situ hybridization (FISH) metabolic oligosaccharide engineering and bioorthogonal click chemistry (BCC) combined with whole-body imaging to monitor host-microbiota interactions. Research shows that when the mucosal surface is damaged, the microenvironment and resident microbiota of the original tissue will be rebuilt, but if not repaired in time, the damage will continue to expand, eventually leading to recurring inflammation, thereby inducing inflammation and cancer. By utilizing tumor-derived carbon sources and other nutrients, the microbiota present on or within

the tumor surface may coexist with the tumor's immune microenvironment for a long time [35]. Recurrent mucosal inflammation caused by microorganisms is one of the causes of cancer. In addition, the body's cellular immunity caused by microorganisms has also attracted certain attention. Chang et al. studied the molecular biological mechanism of microbial-mediated cellular immunity leading to lung cancer, and have demonstrated the link between chronic obstructive pulmonary disease (COPD) and lung cancer in population-based studies. demonstrated the critical role of Th17 cell- mediated inflammation in lung tumorigenesis [36]. C. Jungnickel et al. conducted in- depth research on IL-17C in the microenvironment of lung cancer. They found that IL- 17C in epithelial tissue can stimulate neutrophils to produce an inflammatory response, and further revealed that this phenomenon can regulate the inflammatory response in the lung cancer microenvironment. Abnormal microbiota in COPD patients are associated with accelerated tumor development [37]. To test whether there is a clear link between allergic airway inflammation and lung cancer, CE Ochoa et al. used mouse sensitization experiments to find that eosinophilic lung inflammation is associated with increased levels of helper T-cell factor 2. and mucinous metaplasia of the airway epithelium, similar to that seen in patients with asthma. This study shows that the nature of inflammation is clearly specific in the promotion process of lung cancer, and IL-6 plays an important role in the promotion process of lung cancer [38]. Overstimulation of the innate immune system can make the immune system less sensitive to pathogens and produce an adaptive response. As tumors develop and develop, they express anti-inflammatory molecules and create a microenvironment that suppresses immune responses. If we can build a microenvironment rich in T cells and M2 macrophages, we can express programmed death ligand 1 (PD-L1), bacterial toxic T- lymphobacteria associated protein 4 (CTLA4) or transforming growth factor  $\text{tgf-}\beta$ . These factors suppress antitumor natural killer (NK) lymphocyte responses and promote tolerance and tumor immune evasion. Lung microbiota dysbiosis and the presence of specific bacterial strains may contribute to the generation of this immunosuppressive microenvironment.

### 3.3 Create an inflammatory environment

Microorganisms (lactobacilli, bifidobacteria, etc.) or their products (such as cellulose decomposition products short-chain fatty acids, amino acid metabolite peptides, carbamide, phenylalanine, fatty acid metabolites, etc.) enter through the mucosal barrier from the gastrointestinal tract. The blood thereby mediates inflammation. IFN- $\gamma$ , as a gastrointestinal metabolic factor, is the main cytokine in the pathogenesis of inflammatory bowel disease. Langer et al. analyzed the vascular-directed pathogenic function of IFN- $\gamma$  and found that it drives the pathogenesis of inflammatory bowel disease by destroying the vascular barrier. IFN- $\gamma$  exerts a pathogenic effect by destroying the adhesion junction protein VE-cadherin and causing the destruction of the vascular barrier. Intestinal vascular barrier dysfunction has also been demonstrated in patients with inflammatory bowel disease

(No suitable example found) [40]. Pulmonary inflammation can trigger systemic innate responses that increase the susceptibility of the intestine to inflammatory attack [41]. Pulmonary inflammation and gut microbes interact through the gut-lung axis. The composition of the gut microbiota is determined by the local habitat, which itself is shaped by immune stress, such as mucosal IgA. Using a mouse model of a restricted antibody repertoire, Tomasz et al. determined the role of antibody-microbe interactions in the formation of bacterial communities with enhanced L-tyrosine metabolism. This model results in increased concentrations of p-cresol sulfate (PCS), which protects the host from allergic airway inflammation. PCS selectively reduces CCL20 production by airway epithelial cells due to the uncoupling of epidermal growth factor receptor (EGFR) and Toll-like receptor 4 (TLR4) signaling [42]. The molecular mechanism of inflammation in tumor occurrence and development can be summarized into two parts: Caused by tumors. Reactive oxygen species (ROS) and reactive nitrogen intermediates (RNI) produced by inflammatory cells may cause mutations in adjacent epithelial cells. At the same time, these inflammatory cells can also produce cytokines that stimulate ROS and RNI in precancerous cells.

Furthermore, epigenetic modifications that are beneficial in promoting tumor formation may occur under inflammatory conditions. The inflammatory response associated with tumors will enhance the ability to generate substances such as reactive oxygen species, reactive nitrogen species, and cytokines. Waqar et al., evaluating the response of patients with c-MET-positive squamous cell carcinoma (SCC) to telisotuzumab vedotin, an antibody-drug conjugate targeting c-MET, found that cases of pneumonia were observed in some SCC patients. , this study showed that pneumonitis was an unexpected toxicity observed in patients with SCC [43]. Myers et al. found that lung adenocarcinomas often evolve from mucosal glands, occur around the lungs, and in many cases can be found in scars or areas of chronic inflammation [44]. Tumor promotion. Cytokines produced by tumor-infiltrating immune cells can activate important transcription factors in premalignant cells, such as NF- $\kappa$ B or STAT3, which regulate many processes that promote tumor development, including survival, proliferation, growth, vascularization, and invasion. As part of a positive feed-forward loop, NF- $\kappa$ B and STAT3 induce the production of chemokines that attract additional immune/inflammatory cells to sustain tumor-associated inflammation. Patients with non-small cell lung cancer (NSCLC) may develop pulmonary inflammation leading to pulmonary toxicity after radiotherapy (RT). Siva et al. studied the dynamics of plasma inflammatory cytokines induced by RT in patients with non-small cell lung cancer (NSCLC). Determining clinical predictors of toxicity. This study demonstrates the induction of inflammatory cytokines during and after radiotherapy in patients with non-small cell lung cancer. Early changes in IP-10, MCP-1, eotaxin, IL-6, and TIMP-1 levels are associated with higher levels of toxicity [45]. Measuring cytokine concentrations during radiation therapy can help predict lung toxicity and lead to new treatment strategies. Kidd et al. found that patients with hypermetabolic non-small cell lung cancer who lost weight also had elevated levels of inflammatory mediators, such as acute phase proteins. Increased lymphocyte ratio (NLR) also led to poor prognosis in patients with small cell lung cancer, and preoperative acute phase C- Elevated CRP is associated with poor prognosis after resection of non-small cell lung cancer [46].

### 3.4 Affect metabolism

The dynamic balance of the microbiome is closely related to the host metabolism, and this balance significantly affects the host metabolism. Due to various factors, the dynamic balance of the microbiome is disrupted, causing toxic metabolites to accumulate in the lungs. As harmful substances continue to accumulate, they eventually cause lung tumors and lung cancer. Using gas chromatography-liquid

chromatography and 16srRNA gene sequencing, Keren et al. found that patients with gallstones had higher overall fecal bile acid concentrations, reduced microbial diversity, fewer beneficial Roseola species, and an abundance of uncultured Oscillatoria spirillum species. . Research results revealed that microbial changes are associated with the formation of acetaldehyde and deoxycholic acid. Deoxycholic acid, as a metabolite of bile salts, is considered to be directly related to esophageal cancer and liver cancer [47]. Microorganisms can also directly produce some metabolites that lead to the occurrence of cancer. The fermentation of microorganisms can lead to the production of adenosine. Adenosine is an effective signaling molecule that has an impact on lung diseases. Continued adenosine exposure can damage the lung endothelial barrier. of integrity, Gao et al. induced endogenous apoptosis 1/2- mediated intracellular adenosine signaling by balancing nucleoside transporters. demonstrated that activation of p38 and c-Jun N-terminal kinase contributes to sustained adenosine-induced mitochondrial reactive oxygen species production. Sustained adenosine exposure promoted mitochondrial fission and increased mitophagy. Finally, mitochondria-targeted antioxidants prevented mitochondrial fission induced by sustained adenosine exposure and improved cell survival. Results suggest that inhibition of balanced nucleoside transporters by 1/2 mitochondria-targeted antioxidants may be a potential treatment for lung diseases associated with persistently elevated adenosine [48]. The imbalance of the microbiome will lead to an increase in pathogenic factors and affect the normal metabolism of



the host. This is one of the important mechanisms leading to the occurrence of lung cancer that deserves attention.

### 3.5 Genetic toxicity

Certain bacterial molecules can induce DNA damage and produce genotoxicity. That is, it causes cell death and affects the expression of tumor suppressor genes and oncogenes. DNA damage caused by the cells themselves can promote mutations and cancer, and an increasing number of genes promote DNA damage through various direct and indirect mechanisms. After identifying the preferred set of candidate genes using colocalization analysis, Byun J and his team further investigated them using cell- based DNA damage detection methods. They individually reduced the expression of these candidate genes and observed their effects to model changes in allelic expression patterns associated with lung cancer. They then assessed the extent of DNA damage caused by this manipulation. Among these 19 genes, seven were shown to be overexpressed (including IRF4, AK9, CYP21A2, DCBLD1, SECISBP2L, CCDC97, and FUBP1), while the other two were downregulated (PPIL6 and ACTR2, respectively). Among them, IRF4 has attracted attention due to its intrinsic DNA damage-enhancing effect in lung cells due to its higher levels in lung tissue, which may be one of the key factors driving the risk of developing lung cancer. This study expanded the list of genes associated with lung cancer DNA damage and assigned a known cancer-promoting phenotype (DNA damage) to many lung cancer risk genes [49]. In addition, chemicals produced by bacteria, such as reactive oxygen species produced by *Porphyromonas*, hydrogen sulfide produced by *Clostridium cholophilum*, and superoxide dismutase produced by various bacteria, can cause genome instability and increase the probability of cancer. . In addition, microorganisms may also exert carcinogenic effects by integrating their DNA into the host. HBV (hepatitis B virus) enters different stages of development, including integration of HBV-DNA, clonal expansion of liver cells, production of HBV antigens, and specific immune responses against HBV. According to the research results of Mason et al., they found that all patients with chronic HBV infection had a large number of HBV-DNA integrations in the genome, and the amount of clonal hepatocyte expansion exceeded the expected level [50]. Chen et al. studied the transcriptional signals of novel coronavirus infection and found

that the host transcriptional response of lung adenocarcinoma cells to novel coronavirus infection is broadly similar to the host response to multiple viruses in different model systems and patient samples [51] . Shared genes and pathways that are part of the viral host response point to possible therapeutic strategies for novel coronavirus and cancer.

## 4. Detection method

### 4.1 16srRNA gene sequencing technology:

The development of 16srRNA gene sequencing technology has greatly improved the ability to detect bacterial flora involved in lung cancer carcinogenesis inscientific research and clinical practice. Since the base sequences of the 16srRNA gene fragments of different types of bacteria are different, by designing PCRprimers, decoding, and amplifying the DNA samples extracted from the collectedmicroorganisms, the required 16srRNA fragments can be obtained. According to these base sequences The difference determines the number and type of bacterial flora under study. The advantages of 16srRNA gene sequencing technology such as high throughput, low cost, and high efficiency make it widely used in bacterial detection, tumor diagnosis, cancer prevention, etc. In recent research, this technology has been widely used to detect and analyze microbial species and populations. In recent research, this technology has been widely used to detect and analyze microbial species and quantities. Liu's recent research shows that this technology has been widely used to identify and count microbial species. Forexample, a study by Liu et al. used 16srRNA gene sequencing technology to explore the development trend of the microecology of chronic sinusitis [52]. By observing the rich and diverse bacterial populations in the sinuses and based on

their functional properties, they provide useful reference information for immunotherapy of chronic sinusitis. Xu et al. used 16s rRNA gene sequencing technology to study composite biomarkers of autism spectrum disorder. By detecting the microbiota of stool samples of patients with autism spectrum disorder, they showed that there is a relationship between the intestinal microbiota and patients with autism spectrum disorder. The connection provides a new way to use microbial biomarkers to diagnose patients with autism spectrum disorder [53]. Therefore, we should make full use of the advantages of 16s rRNA gene sequencing technology and widely apply this technology to various fields, in order to minimize the pain of patients and improve the quality of life.

#### 4.2 Illumina Genome Sequencing Technology:

Illumina genome sequencing technology is often used in conjunction with 16s rRNA gene sequencing technology for bacterial flora analysis. There are significant differences in the hypervariable regions of 16s rRNA base sequences between different strains. Usually, during bacterial flora analysis, only the hypervariable regions of the obtained 16s rRNA base sequences are sequenced and compared to distinguish different bacterial groups. Illumina genome sequencing technology can detect short (75-250) gene fragment sequences [54]. The length of the 16s rRNA hypervariable region is about 30-100 base pairs, so Illumina genome sequencing technology usually only needs to be applied once to complete it. Sequence analysis of the hypervariable region of 16s rRNA. MW Gray et al. used Illumina genome sequencing technology to analyze sequences corresponding to the highly conserved secondary structure core

prevalent in small subunit rRNA, explored the evolutionary origins of organisms and organelles, and provided methods for the study of global phylogeny [55]. The low-cost Illumina genome sequencing technology and its adaptability to 16s rRNA gene sequence detection make this technology more widely applicable.

#### 4.3 FISH technology:

FISH technology is an important non-radioactive in situ hybridization technology and is currently widely used in many fields such as chromosome research, gene mapping, and tumor diagnosis. We focused on the application of FISH technology in microbiota research. Design oligonucleotide primers based on the base sequence of the 16s rRNA of the target microorganism, usually 16s rDNA primers. By adding a reporter molecule and fluorescein to the primer, an RNA-DNA double-stranded complex can be generated after a process of denaturation, annealing, and recombination. Then on the fluorescence detection equipment, we can conduct qualitative, quantitative and localization research on the targeted microorganisms [56]. Thanit et al. used peptide nucleic acid fluorescence in situ hybridization (PNA-FISH) in their experiments to identify bacteria from nasal polyps or sinus tissue in samples from sinus patients. Mucosal presence of *Staphylococcus aureus* was found to be associated with IL-5 and SE-IgE-positive chronic rhinosinusitis with nasal polyps and nasal polyps from patients with cystic fibrosis. Chronic rhinosinusitis is associated with the presence of *Pseudomonas aeruginosa* in the mucosa [57]. It provides clues for the study of the relationship between mucosal microorganisms and inflammation in chronic rhinosinusitis. Although FISH technology has played a key role in disease prevention and treatment, it still has some shortcomings. Since some test objects themselves produce fluorescence, the specificity of FISH technology is reduced and interferes with the test results. Moreover, since FISH technology can only be used to detect microorganisms with known sequences, the number of primers that can be designed is small, resulting in a relatively limited research scope. Usually FISH technology needs to be used in conjunction with other technologies to achieve the desired goal.

## 5. Treatment method

Currently, the treatment method for lung cancer is to integrate multidisciplinary comprehensive treatment with individualized combined treatment. Commonly used treatments include surgery, chemotherapy, radiotherapy, molecular targeted therapy, etc. Depending on the patient's cancer pathological tissue type, clinical grade and other conditions, the best treatment plan will also be different. (1) Radiotherapy and chemotherapy: This type of method is currently the most commonly used clinical cancer treatment method. About 70% of cancer patients require radiotherapy and chemotherapy. By implementing radiotherapy and chemotherapy, we can eliminate or cure local primary cancers or metastatic lesions, thereby controlling the progression of cancer and improving patients' quality of life and extending their lifespan. Because patients with small cell lung cancer are more sensitive to radiotherapy and chemotherapy, radiotherapy and chemotherapy have become the best methods to treat small cell lung cancer. Most patients with extensive-stage small cell lung cancer (ES-SCLC) will develop persistent intrathoracic disease after receiving chemotherapy and prophylactic cranial irradiation. Slotman et al. performed thoracic radiotherapy on some of the subjects and the results showed that all patients with chemotherapy-responsiveness. For patients with responding ES-SCLC, in addition to prophylactic cranial radiotherapy, chest radiotherapy should also

be considered [58]. Bogart et al. studied the treatment of radiotherapy for limited-stage small cell lung cancer and found that long-term treatment can induce tolerance, so in clinical practice, most patients should receive higher doses of once-daily radiotherapy. This study helps guide the selection of thoracic radiotherapy regimens for patients with limited-stage small cell lung cancer and reduces the frequency of adverse events [59]. (2) Surgical treatment: Surgical therapy is often used in the treatment of advanced cancer. It removes part of the tumor to reduce damage to surrounding organs or tissues, thereby achieving the goal of relieving the condition, such as fistula surgery, digestive tract short circuit and other surgical methods. Surgical resection is the preferred method for patients with early-stage non-small cell lung cancer. Surgical treatment can cure some patients with early-stage non-small cell lung cancer, so for these patients, surgery is the best treatment method. Xu et al. studied the efficacy of cryoablation in the treatment of non-small cell lung cancer. The results showed that the recurrence rate of non-small cell lung cancer patients treated with cryoablation was significantly reduced, and the prognosis and life span were relatively good [60]. Wei et al. conducted an in-depth study of patients with non-small cell lung cancer who underwent thoracoscopic lobectomy. Their results showed that ligating the veins first during the surgery can help reduce the spread of tumor cells and improve the survival rate of non-small cell lung cancer patients' survival status [61]. (3) Immunotherapy and targeted drug therapy: Immunotherapy and targeted drug therapy are currently the most ideal and effective methods for treating cancer. Anti-cancer therapies include immunotherapy and targeted drug therapy, which use various approaches to fight cancer. These methods mainly rely on biologically active chemical components or drugs to assist the immune system in destroying cancer cells, curbing their ability to reproduce, and preventing signaling related to the formation of new blood vessels; at the same time, they can also deliver toxic substances to the interior of cancer cells. Trigger tumor cells to self-destruct, or reduce levels of hormones necessary for cancer cell proliferation. Patients with advanced lung cancer are in very poor physical condition and cannot tolerate radiotherapy or chemotherapy. They are no longer able to undergo surgery. Molecular targeted drug therapy and immunotherapy can be used. Awad et al. used personalized neoantigen vaccine NEO-PV-01 combined with pemetrexed, carboplatin, and pembrolizumab to treat patients with advanced non-squamous non-small cell lung cancer. The results showed that this regimen was effective in advanced non-squamous non-small cell lung cancer. It is safe and highly effective in the treatment of patients with non-small cell lung cancer [62]. In recent years, immune checkpoint inhibitors (ICIs) have been used as new therapeutic methods for molecularly targeted drugs to treat cancer due to their effectiveness and safety [63]. At present, this treatment method has been widely used in the treatment of various solid tumors. Some bacteria such as Bifidobacteria, Bacteroides

*thetaiotaomicron* and *Bacteroides fragilis* in the genus *Bacteroides* have the effect of enhancing anti-tumor immunity and enhance the effect of ICIs. Dizman et al. studied the impact of bifidobacteria on the response to checkpoint inhibitors in cancer patients. These data show that bifidobacteria appear to enhance clinical outcomes in patients with metastatic renal cell carcinoma treated with nivolumab-ipilimumab. , the disease-free survival period was significantly prolonged, and the response rate was also improved [64]. In addition, long-term use of ICIs will produce

certain side effects, which have been observed including arrhythmia, pericarditis, myocarditis, hypothyroidism, joint pain, etc. Tawbi et al studied the antitumor efficacy of relatlimab (a LAG-3-blocking antibody) and nivolumab (a PD-1-blocking antibody) in previously treated melanoma patients and showed an 18.9% Patients experienced grade 3 or 4 treatment-related adverse events [65]. In addition, long-term use of ICIs will develop drug resistance. In the treatment of advanced lung cancer, immunotherapy and targeted drug therapy are the main methods. Since both methods have high safety and efficacy, they have been widely used in clinical practice. The emergence of immune checkpoint inhibitors has further enhanced patients' survival, alleviated patients' pain and had fewer side effects. However, in recent years, with the widespread application of immune checkpoint inhibitors, the occurrence of related adverse events and drug resistance has attracted certain attention. How to further improve the application of immune checkpoint inhibitors requires more in-depth research.

## 6. Conclusion

This review summarizes the relationship between the microbiota and lung cancer. Lung microbes originate from multiple pathways and are common in tumor tissue. In addition to the lung microbiota, the microbiota in multiple tissues and organs of the body are significantly related to the occurrence of lung cancer. The microbiota in different tissues and organs are different, but most of them are characterized by a decrease in probiotics and an increase in pathogenic bacteria. , the commonality is that the microbiota of lung cancer patients often exhibits a loss of diversity, a reduction in total bacterial abundance, and changes in bacterial composition. The mechanism by which the microbiota affects the occurrence of lung cancer is complex. Microbiota imbalance, mediating immune responses, creating an inflammatory environment, affecting metabolism, and inducing genotoxicity are all important pathways leading to the occurrence of lung cancer. Already relatively mature sequencing technologies such as 16srRNA gene sequencing technology, Illumina genome sequencing technology, and FISH technology provide important methods for the determination of microbiota. The widespread application of these technologies allows the study of the relationship between microorganisms and cancer, increases the basis for cancer diagnosis, and improves cancer diagnosis. therapeutic effects are possible. Currently, surgical therapy, chemotherapy, radiotherapy and molecular targeted therapy are the mainstream methods of tumor prevention and treatment. We can choose the most suitable method for different lung cancers to minimize the patient's suffering and achieve the goal of cure. The relationship between the microbiome and lung cancer is complex and changeable. With further research, more new diagnostic and treatment methods will be applied to the clinical practice of lung cancer. The pain of lung cancer patients will be alleviated and the quality of life will be improved.