# New Paradigm of Gastric Pathogenesis: The Important Role of Gastric Microbiota

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## ABSTRACT

*Microbiota was deemed essential as it involved in energy metabolism, nutrient absorption, intestinal immune system maturation, and pathogen protection. Gastrointestinal microbiome played essential roles in human body, such as immune response regulation, pathogen colonization, and few other diseases. The relation between gastric microbiota and host were difficult to explore for years due to unculturable microbes. Stomach with its acid production was presumed to be sterile and unfavorable for bacterial growth until the discovery of Helicobacter pylori (H. pylori). It dominates the stomach as it was estimated to colonize almost 50% global population. H. pylori infection was linked to the development of chronic gastritis and recognized as a definite carcinogen. There was a probability that the alteration of gastric microbiota likely influenced gastric immunobiology and possible gastric diseases. Recent studies showed that five phyla consist of Firmicutes, Bacteroidetes, Actinobacteria, Fusobacteria and Proteobacteria have been discovered in stomach mucosa which might contribute to the pathological process. In addition, genera such as Lactobacillus, Escherichia-Shigella, Lachnospiraceae, Burkholderia, and Nitrospirae were considered to have a role on gastric carcinogenesis.*

***Keywords:*** *microbiota, gastric diseases, H. pylori, stomach, cancer, infectious disease*

## ABSTRAK

*Mikrobiota dianggap esensial karena perannya dalam metabolisme energi, absorpsi nutrisi, maturasi sistem imun intestinal, dan proteksi terhadap patogen. Mikrobiota gastrointestinal memegang peran penting dalam tubuh seperti regulasi respons imun, kolonisasi patogen dan penyakit-penyakit lain. Hubungan antara microbiota lambung dan pejamu sulit diketahui karena sulitnya kultur mikroba ini. Lambung dengan produksi asamnya*

*diperkirakan memiliki lingkungan yang steril hingga ditemukannya Helicobacter pylori (H. pylori). Bakteri ini mendominasi gaster serta diperkirakan sudah mengkolonisasi hampir separuh penduduk dunia. Infeksi H. pylori banyak dikaitkan dengan perkembangan gastritis kronis serta dikategorikan sebagai karsinogen. Perubahan dari mikrobiota lambung ini diperkirakan dapat memengaruhi imunobiologis pada lambung dan kemungkinan menimbulkan penyakit lambung. Penelitian terkini menunjukkan lima filum yang terdiri dari Firmicutes, Bacteroidetes, Actinobacteria, Fusobacteria dan Proteobacteria, yang ditemukan di mukosa lambung dan mungkin dapat berkontribusi terhadap proses patologis ini. Ditambah dengan beberapa genera seperti Lactobacillus, Escherichia-Shigella, Lachnospiraceae, Burkholderia, dan Nitrospirae juga diduga memiliki peran dalam karsinogenesis lambung.*

***Kata kunci:*** *mikrobiota, penyakit lambung, H. pylori, lambung, kanker, penyakit infeksi*

## INTRODUCTION

Microbiota was deemed essential as it involved in energy metabolism, nutrient absorption, intestinal immune system maturation, and pathogen protection. Gastrointestinal microbiome played essential roles in human body, such as immune response regulation, pathogen colonization, and few other diseases.[1] Alteration to this harmony could predispose to cancer.[2] Human stomach was thought to be sterile due to its high acidity up until a legion of microbiota was found there. Thickness of mucus layer and gastric peristaltic movement were another factor to withhold bacterial growth. *Actinobacteria, Proteobacteria, Firmicutes, Fusobacteria* and *Bacteroidetes* were the five major phyla discovered here, nevertheless.[3–5] The composition of gastric microbiota changed by time.[3] The species of *Helicobacter pylori* dominates as it was estimated to colonize almost 50% global population.[4,6] However, only small amount of these individuals develops gastric cancer.[6,7]

The relation between other gastric microbiota and host were difficult to explore for years due to unculturable microbes. The hypochloridria condition in atrophic gastritis were thought to allow growth of another microbes.[6] Evidence showed that these organisms play a greater role in disease progression. Studies shown prospective connection of microbes other than *H. pylori* in carcinogenesis and even another gastroduodenal diseases.[1,8] Hence understanding the interaction between microbiota and possible risk of disease is necessary.

### Gastric microbiota composition

Stomach with its acid production was deemed sterile and unfavorable for bacterial growth.[4] Added by another factors, research does not seem to uncover the various population of microbiome, prior to the discovery of *Campylobacter pyloridis*, now known as *Helicobacter pylori* in 1982. The gram-negative bacteria produce urease enzyme, in which produces ammonia, making it able to neutralizes acid and colonize gastric epithelium. This bacteria had significant impact both on duodenal and oral region.[3] *Helicobacter pylori* had known to infect 50% of world population, but only small amount suffer from diseases. When present, it is the dominating microbiota in stomach. While *H. pylori* infection alters microbial composition by increasing stomach's pH and create environment suitable for bacterial colonization, a research found that the absence of *H. pylori* showed more diverse gastric microbiota instead. *Actinobacteria, Firmicutes* and *Bacteroidetes* were exuberant in *H. pylori* negative environment.[1,5]

The interaction in between these bacteria and host is not yet conclusive. Moreover, the biodiversity of gastric microbiota remained inexplicable since most of it are unculturable. Hence, another approaches have been used to detect these microbiota.[1,6] Sequencing techniques used had revealed main genera found in stomach, being *Streptococcus, Rothia, Veillonella*and *Prevotella.* There were even differences of microbiota composition in gastric fluid and mucosa. *Actinobacteria, Bacteroides* and *Firmicutes* were the main phyla found in gastric fluid while *Firmicutes* and *Proteobacteria* prevailed in gastric mucosal samples.[1] In addition, few transient bacteria had been found without colonizing gastric mucosa. Researchers had found another bacteria that resistant to acid, such as *Streptococcus, Neisseria* and *Lactobacillus*, which all came from either oral cavity or duodenum.[3] It was revealed that more than 600 bacterial phylotypes are active bacterial community in gastric mucosa.[9]

The shifting arrangement of gastric microbiota is affected by diet, medication, gastric mucosal inflammation and *H. pylori* colonization.[1] Prolonged usage of proton pump inhibitors (PPIs) and H2-

antagonists affect its growth since bacteria will flourish at pH of more than 3.8. Oro-pharyngeal and fecal-like bacteria was found more in PPIs patients. Sustained infection by *H. pylori* results in the destruction of gastric epithelial cells, hence increasing its pH and promote colonization of transient bacteria.[3] Research had reported increased *Spirochaeta, Streptococcus, Lactobacillus* and *Veillonella* in environment with increased pH.[4] Moreover, ammonia and bicarbonate produced were used as substrate by other bacteria.[3]

### Gastric microbiota and diseases

The recognition of *H. pylori* as a definite carcinogen by World Health Organization (WHO) was in 1994 since it induces chronic gastritis and finally, gastric cancer. It occurred by progressing from atrophic gastritis, intestinal metaplasia, to gastric cancer.[6] Several virulence factors, host's genetic, and supporting condition have all contributed to this.[9] In the absence of antibiotic therapy, *H. pylori* infection remained, while first exposure usually occurred in childhood. The infection generates gastritis (antrum-predominant) by rising acidity in antrum and atrophic gastritis in corpus, which eventually leads to gastric cancer.[4] Atrophic gastritis happens due to continuous inflammation in gastric mucosa and destruction of hydrochloric acid secreting glands.[6] Early diagnosis could lower mortality as atrophic gastritis is an influential pre-cancerous disease. In addition, *H. pylori* infection was also related to peptic ulcers, mucosa-associated lymphoid tissue lymphoma (MALT lymphoma) and non-cardia gastric adenocarcinoma.[1]

Microbes accompanying *H. pylori* has been proposed to alter infection's outcome. Despite the independence between microbial composition of *H. pylori* contribution and relative abundance, serological status was associated with reorganization in microbial composition.[10] The existence of *H. pylori* in stomach caused decreased bacterial divergence; which could be seen as the amount of *Treponema, Prevotella,* and *Tannerella* in *H. pylori*-induced atrophic gastritis are lower.[1] In gastric cancer patients, culture analysis showed more microorganisms than those without. These patients had a myriad microbes ranging from *Streptococcus, Lactobacillus, Veillonella* and *Prevotella* with decreased *H. pylori* strain.[3]

Microbiome imbalances were linked to various diseases including cancer. There are conflicting results regarding gastric microbiome composition during carcinogenesis, in terms of its diversity and abundance.[8] The disequilibrium of microbial community in gastric mucosa could increase cancer's risk as it was observed in a research involving 54 gastric cancer patients and 81 with chronic gastritis, through 16s rRNA gene profiling, patients with gastric cancer had less microbial heterogeneity.[11] In line with this, a research found gradual decreased bacterial diversity between non-atrophic gastritis, intestinal metaplasia and gastric cancer.[12] Few of dominant phyla in gastric cancer were; *Actinobacteria, Proteobacteria, Firmicutes, Fusobacteria* and *Bacteroidetes*.[1] In addition, gastric cancer patients were found to have more non-*Helicobacter* Proteobacteria, while Hsieh et al reported *Lactobacillus, Fusobacterium* and *Clostridium* were regularly found in gastric cancer. Moreover, in another research involving bigger sample of 315 patients (212 with chronic gastritis and 103 with gastric cancer), it was found an increased bacterial load per gram tissue in cancer group. From this, five genera of *Lachnospiraceae, Lactobacillus, Nitrospirae, Burkholderia* and *Escherichia-Shigella* were found, all with a possibility to induce cancer.[2] The loss of parietal cells demonstrate the loss of defense in these lesions.[9] Nonetheless, bacterial cornucopia in stomach had long found in precancerous settings.[8,11,13]

Gastric bacteria other than *H. pylori* involved in gastric cancer were lactic acid bacteria (LAB); consist of *Lactobacillus, Streptococcus, Lactococcus* and *Bifidobacterium.* LAB could increase N-nitroso compound that promotes angiogenesis and proto-oncogene expression while inhibit apoptosis. It could also induce multipotency in cells, give way to tumor development, and increase production of exogenous lactate. Increased concentration provides source for oxidative cancer cells, hence, contribute to cell migration. It influences the immune response by inhibiting the function and survival of T and natural killer cells.[5] *Lactobacillus* a gram-positive bacteria, had been found to be able to inhibit *H. pylori* adhesion to mucosal cells and inhibit urease activity. It has been used as probiotics to prevent infection, but it was also able to induce inflammation to epithelial cells. However, *Lactobacillus* could increase the level of lactic acid, which could be used as energy source for tumor cells, encourage inflammation and provoke

tumor angiogenesis.[10] *Escherichia- Shigella* was found increased in gastric cancer and colorectal cancer.[2] Few members of *Nitrospirae* phylum were recognized to take part in nitrites and nitrates metabolism in which it is a risk factor for gastric cancer development.[14] Nitrite is a precursor for n-nitroso compounds. Bacteria could convert nitrite to nitrosamine, which play a role in the development in atrophic gastritis and a renown carcinogen that develops into gastric cancer. *Staphylococcus*, *Clostridium, Veillonella,* and *Haemaphilus* may add to the formation of N-nitroso compounds.[1] Another microbes of *Lachnospiraceae* family were found to be increased in gastric cancer patients as it was proposed to influence the inflammatory process.[6] Moreover, few oral bacteria were found abundant in gastric cancer stage.[8]

Other bacteria than *H. pylori* that has possible roles in gastric carcinogenesis have been observed in experimental animal models.[9] Researchers had performed studies on insulin-gastrin transgenic (INS-GAS) mouse model to analyze the association between microbiota and gastric cancer.[15] Through manipulating the microbial composition in INS-GAS mice with three variations; complex, germ-free, and limited microbiota (only *Bacteroides, Lactobacillus* and *Clostridium)*, they found that both complex and limited gastric microbiota were linked to the similar rate of gastric carcinogenesis. However, when compared between germ-free and restricted microbiota INS-GAS mice, the later showed more gastric corpus inflammation, epithelial defects, epithelial hyperplasia and dysplasia.[16] *H. pylori* infection was linked to the development of chronic gastritis, but the alteration of gastric microbiota composition also plays a role in carcinogenesis.

## CONCLUSION

Gastric microbiota had an essential part in the development of gastric diseases. Recent studies had shown various bacteria other than *Helicobacter pylori*. Five phyla have been discovered in stomach mucosa, such as *Firmicutes, Bacteroidetes, Actinobacteria, Fusobacteria* and *Proteobacteria* which might contribute to the pathological process. Several genera such as *Streptococcus, Lactobacillus, Bifidobacterium* and *Lactococcus* have been found and correlates to gastric cancer incidence. However, the role of these microbiota has not been clarified yet. Therefore, we believe that further research to understand the detailed mechanism of the interaction between gastric microbiota and its environment is needed to explain the role of non *H. pylori* bacteria to allow new viewpoint for prevention and treatment of gastric diseases in the future.

### Conflict of Interest

The authors declare no conflict of interest.

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