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Role of *Helicobacter pylori* in development of gastric cancer

A Review Article

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By

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Tables of contents

List of Figures

Abstract

Gastric carcinoma is the fourth most common cancer worldwide, with approximately 930,000 new cases diagnosed each year**.** there are

two major types of gastric carcinoma, the intestinal type, which is associated more commonly with environmental perturbations, and the diffuse type, which is ascribed etiologically more often to host genetic factors**.** *Helicobacter pylori (H.pylori)* is a gram negative microaerophillic bacterium that inhabits various areas of stomach and duodenum. Normally the acidic environment of stomach prevents the survival of viruses, bacteria and other micro-organisms. Initial investigations into a possible association of H pylori with gastric cancer that were conducted in the 1980s and early 1990s provided only weak evidence for a link between H pylori and gastric cancer. But the decline in incident of cancer support this theory. In this review we will discuss the relationship between incident of cancer and the infection.

Keywords: carcinoma, *helicobacter pylori,* stomach

Introduction

Gastric cancer is the fourth most common cancer and the second most common cause of cancer death. The incidence of gastric cancer depends on gender and geographic variation. Men are 2–3 times more sensitive than females. Incidence shows a great deal of geographical diversity. It should be noted that more than 50% of new incidents occur in developing countries. The incidence of gastric cancer has declined in most parts of the world over the past few decades. Sporadic intestinal gastric cancer type declines have been observed, and the incidence of diffuse GC types has increased (1). When considering men and women combined, GC is responsible for 8.2% of cancer deaths worldwide, making it the third most common cause of death from cancer (24).

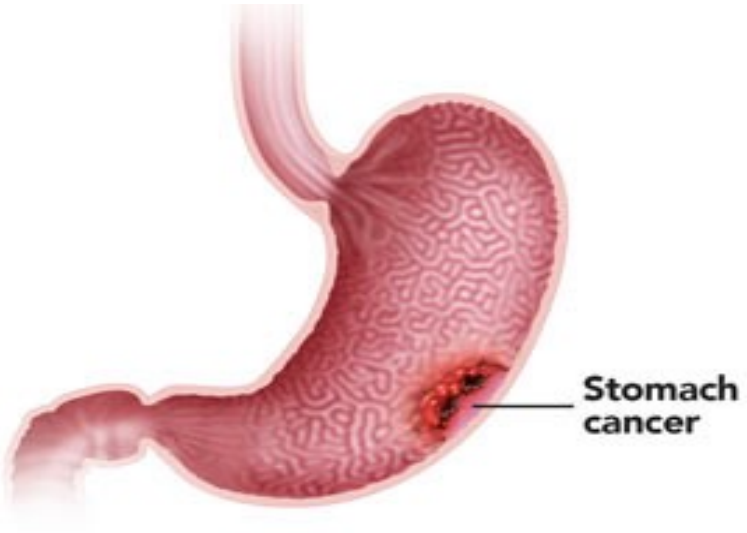

Figure 1. Gastric CA

Histologically, there are two major types of gastric carcinoma, the intestinal type, which is associated more commonly with environmental perturbations, and the diffuse type, which is ascribed etiologically more often to host genetic factors. Intestinal type carcinoma cells show irregular tubular structures, harboring pluri stratification, multiple lumens and reduced stroma, often associated with intestinal metaplasia in neighboring mucosa. Diffuse-type carcinoma cells are poorly differentiated and are characterized by the production of discohesive and secrete mucus, which is delivered into the interstitium. If the mucus remains inside the tumor cell, it pushes the nucleus at the periphery (therefore called signet-ring cell carcinoma). A small portion of diffuse-type gastric carcinomas is of familial origin, caused by mutations in the E-cadherin gene (2).

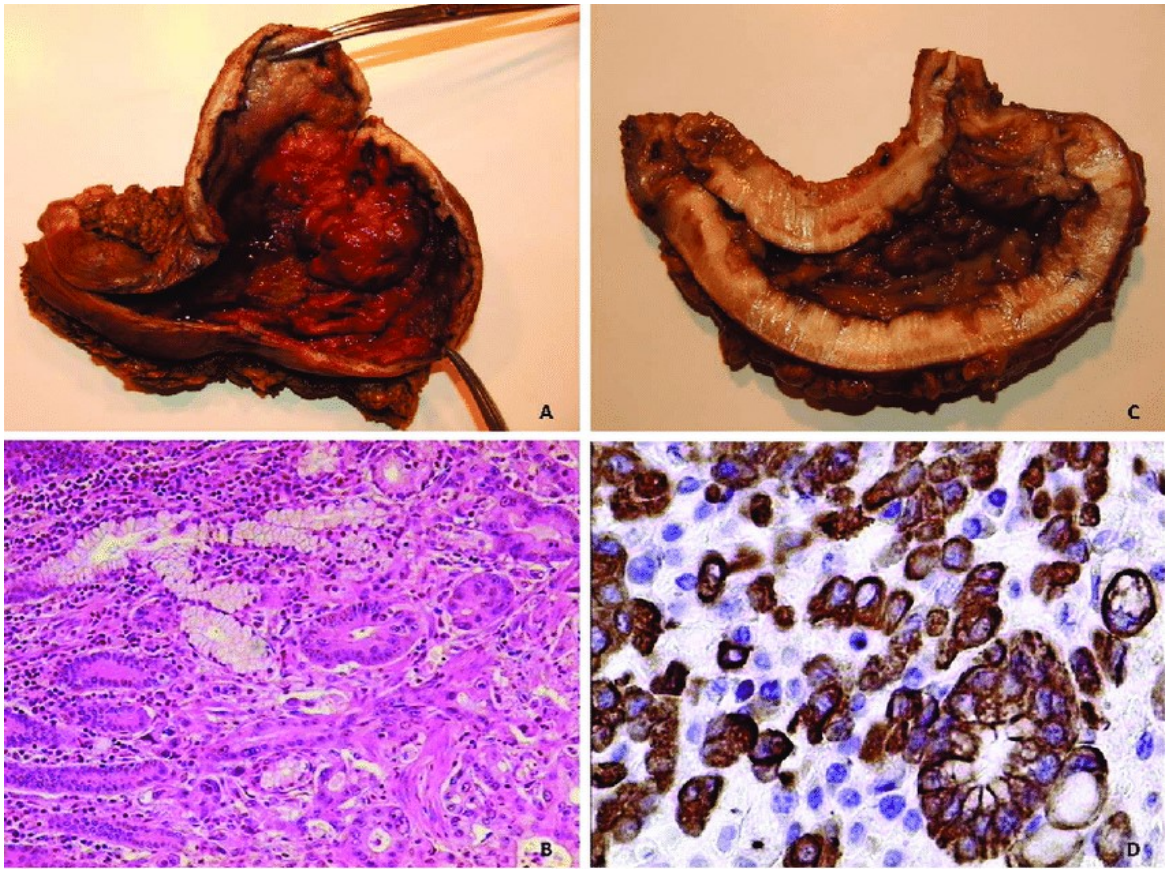

Figure 2. Histological types of Gastric CA: A&B show the intestinal type, C&D show the diffuse type.

Recent epidemiological studies have indicated that Helicobacter pylori plays a key role in the development of both intestinal-type and diffuse-type gastric carcinomas (3). *Helicobacter pylori (H.pylori)* is a gram negative microaerophillic bacterium that inhabits various areas of stomach and duodenum. Normally the acidic environment of stomach prevents the survival of viruses, bacteria and other micro-organisms. However H. pylori have evolved to be uniquely suited to thrive in the harsh stomach environment. The *H. pylori* bacterium secretes urease, a special enzyme that converts urea to ammonia. Ammonia reduces the acidity of stomach, making it more hospitable home for *H.pylori* (4).

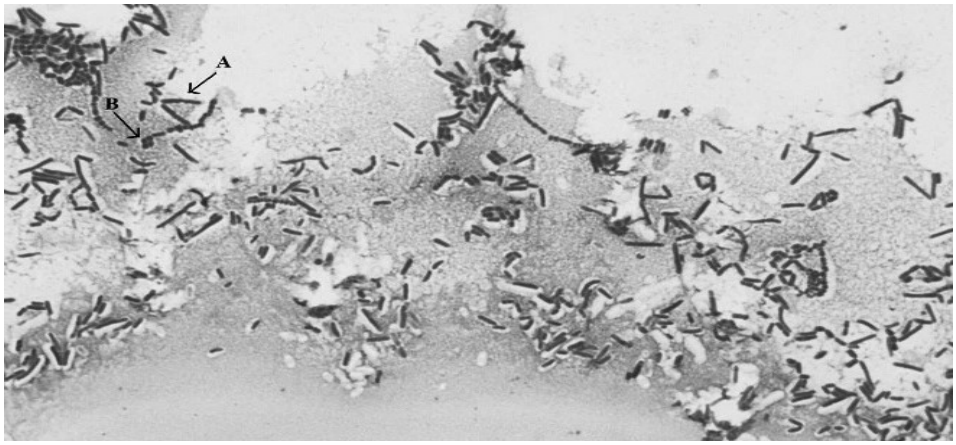

Figure 3. H. pylori under microscope

Helicobacter pylori is a microbial species that specifically colonizes gastric epithelium and it is the most common bacterial infection worldwide. Everyone infected by this organism develops coexisting gastritis, which typically persists for decades, coupling *H. pylori* and its human host into a dynamic and prolonged equilibrium. However, there are biological costs incurred by such long-term relationships. *H. pylori* infection is the strongest known risk factor for malignancies that arise within the stomach, and epidemiological studies have determined that the attributable risk for gastric cancer conferred by *H. pylori* is approximately 75% (5).

Over the last decade, the incidence of gastric cancer has decreased dramatically in some developed countries. This unexpected “triumph” has been attributed to a decreased incidence of *Helicobacter pylori* infection in these populations. Indeed, in 1994, the International Agency for Research on Cancer (World Health Organization) classified *H. pylori* as a definite class 1 carcinogen (6).

In this article, we will discuss the role of *H. pylori* in the incidence and the pathogenesis of gastric carcinoma.

Literature review

H. pylori strains are extremely diverse, freely recombining as panmictic populations. Genetic variability is generated through intra-genomic diversification (for example, point mutations, recombination and slipped-strand mispairing) as well as inter-genomic recombination. The use of broad-range 16S ribosomal RNA (rRNA) PCR coupled with high-throughput sequencing has demonstrated that *H. pylori* does not exist simply as a monoculture within the human stomach but is instead a resident of a distinct gastric microbial ecosystem (7). Initial investigations into a possible association of *H pylori* with gastric cancer that were conducted in the 1980s and early 1990s provided only weak evidence for a link between *H pylori* and gastric cancer. A positive correlation between *H pylori* seroprevalence and gastric cancer in cross-sectional sampling was reported in a study conducted among 13 European nations. However, although some other publications reported similar trends, several others did not find a positive correlation between the presence of *H pylori* antibodies and either gastric cancer or precancerous gastric lesions (8). Around this time, clinicopathologic studies performed to look for evidence of *H pylori* infection (by serum antibodies) or in gastric tissue directly in cases of gastric cancer also yielded inconsistent results. Some studies showed *H pylori* infection rates no higher than those observed in noncancer controls. This is most likely because by the time gastric cancer has developed, extensive intestinal metaplasia and hypochlorhydria had rendered the stomach less hospitable to persistent H pylori colonization, thus also explaining some of the negative findings in the cross-sectional studies discussed earlier (9).

Much more convincing evidence for the role of *H pylori* in gastric cancer came from 3 large cohorts with nested case-controls, in which serum had been banked from cancer-free subjects and the cohort had been followed up for approximately a decade (10-11) In each cohort, evidence of prior *H pylori* infection (evaluated by enzyme-linked immunosorbent assay [ELISA] in the

banked serum) was found to be significantly more common in those subjects who subsequently developed gastric cancer compared with a sample of those who had not Based on the compelling results of these cohort studies. the World Health Organization's International Agency for Research on Cancer declared in 1994 that there was sufficient evidence to classify *H pylori* as a definite (group 1) carcinogen (8).

The role of *H. pylori* infection in the carcinogenic process was first considered to be indirect via the long-term inflammation that is induced. The Th1 type immune response leads to apoptosis of the gastric epithelial cells and to a cell proliferation to compensate for the cell loss. The important production of oxygen free radical species leads to errors during mitosis and an accumulation of mutations. This process may be reinforced by the fact that H. pylori impairs DNA mismatch repair in gastric epithelial cells (12). More recent data have shown that *H. pylori* may also have a direct carcinogenic effect via the CagA protein. CagA interacts with proteins of the tight junctions: ZO-1, JAM and adherent junctions, with E-cadherin leading to a destabilization of these junctions and activation of β-catenin. This effect would be due to the interaction of CagA with the PAR1 kinase (partitioning-defective 1 microtube affinity-regulating kinase MARK) involved in the cytoskeleton structure and cell polarity (13). Other effects of CagA appear after its phosphorylation. It then interacts with the SHP2 phosphatase which inhibits Src dephosphorylation, FAK which regulates the focal adherence plaques to the cellular matrix, and also c-Met, MAPK and other actin cytoskeleton regulators. The outcome is the change in phenotype of the epithelial cell, described as the "hummingbird" phenotype. this change indeed corresponds to an epithelial–mesenchymal transition, with an increase in mesenchymal cell markers and a moderate decrease in epithelial cell markers. Furthermore, these cells express a high level

of CD44, which is a marker of gastric cancer stem cells (CSC) and which present CSC-like properties (14).

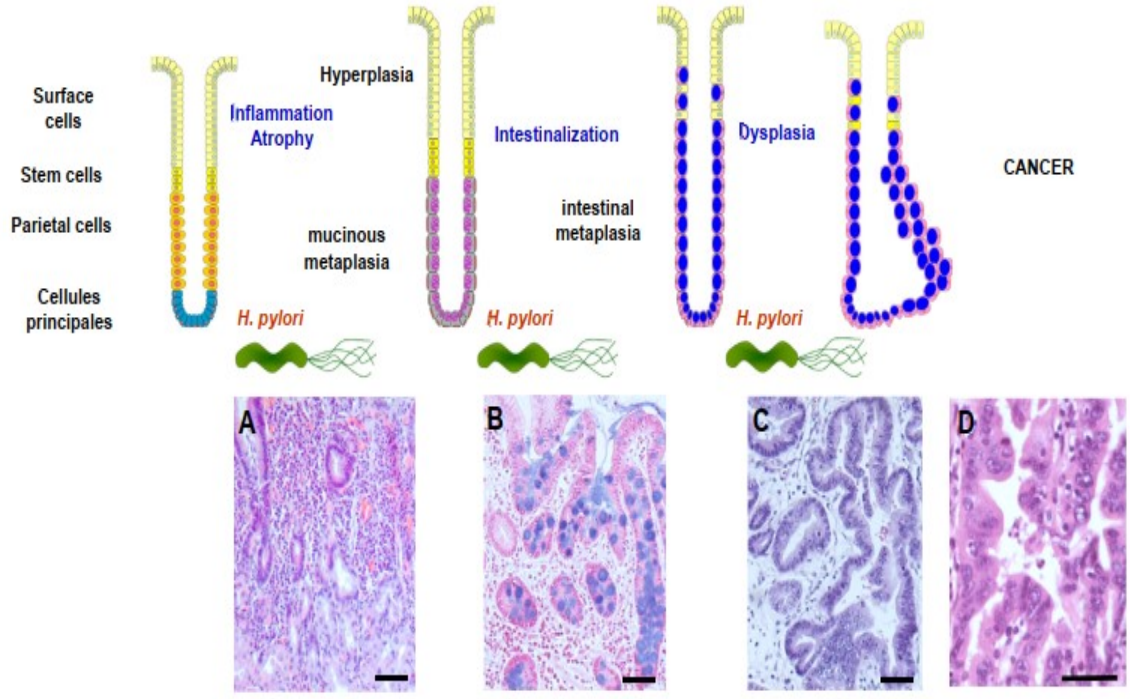

Figure 4. Cascade of histologic changes induced by Helicobacter pylori at level of gastric mucosa.

Activation of Nuclear factor-κB (NF-κB)and up-regulation of IL-8 in gastric epithelial cells were suggested as the critical mechanisms responsible for H. pylori-induced chronic inflammation and gastric carcinogenesis (15). *H. pylori* may activate NF-kB through classical or alternative pathways, depending on the cell type. Activation of NF-kB by *H. pylori* in gastric epithelial cells was mainly through the classical pathway, which is dependent on Cag PAI. In this pathway, CagA can physically associate with and enhance the activity of TAK1, which then activates NF-kB through TRAF6-mediated, Lys 63-linked ubiquitination of TAK1. The role of PI3K signaling in the CagA-induced NF-kB activation and inflammation was supported by the observation that either PI3K inhibition

or small interfering RNA (siRNA) mediated knockdown of NF-kB p65 could suppress H. pylori-induced IL-8 expression (16).

Cancer stem cells (CSCs) represent a subset of cells with the ability to self-renew and differentiate into mature tumor cells. It is now believed that CSCs play a pivotal role in the development of many cancers including gastric cancer. The origin of the gastric CSCs are not completely clear but they have been suggested to stem from the differentiated gastric epithelial cells, local progenitor cells in the gastric mucosa, or bone-marrow-derived cells (BMDCs). Animal studies have shown that the chronic inflammation caused by *H. pylori* in the gastric mucosa could induce the recruitment, homing, differentiation and transformation of BMDCs, suggesting that gastric CSCs may play a role in the H. pylori induced GC. Interestingly, some known pathogenic factors of H. pylori (e.g. CagA protein, T4SS or VacA protein) might not be involved in the mobilization of gastric CSCs, but rather, certain undiscovered virulence factors and cytokines secreted by infected epithelial cells (e.g. TNFa) may contribute instead (17-18).

Inactivation of p53 by mutations has been reported in approximately 40% of gastric cancers, and this is especially common in individuals infected with CagA positive strain of *H. pylori*. It was reported that *H. pylori* could actively alter p53 level in a bimodal fashion in vivo. Infection of H. pylori in Mongolian gerbils for 4–6 h induced an acute accumulation of p53 in gastric mucosa, followed by a relatively low plateau for several weeks, and then a second peak. This dynamic change seems to depend on the balance between the p53 degradation induced by *H. pylori* and intracellular self-defense mechanism. Aberrant activation of oncogenes, DNA damage and a high level of inflammation induced by *H. pylori* could trigger the intrinsic cellular protection mechanisms, which up-regulate the p53 protein (19).

Vacuolating Toxin (VacA) is another virulence factor of *H. pylori* and the *vacA* gene maps to separate loci from the *cagA* on the *H.pylori* chromosome. It functions as an anion channel. All strains of *H. pylori* carry the *vacA* gene unlike the c*ag* island and encodes for the VacA toxin. In mice, VacA induces gastric erosions. In addition to this, it is thought to inhibit the T lymphocyte activation through induction of a G1/S cell cycle arrest, impedance of transcription of IL-2 and downregulation of IL-2 through the targeting the nuclear factor of activated T cells (NFAT) signaling pathways. The exact role of v*acA* in oncogenesis is not yet known. However, strains possessing s1/m1 allele are associated with increased risk of gastric cancer (20). Cyclo-oxygenase-2 (COX-2) Is Induced by Inflammatory Cytokines. COX-2 expression has been demonstrated in cell cultures co-cultured with *H. pylori*. Higher levels are also seen in the gastric mucosa of infected subjects. COX-2 expression is more pronounced in pre- and malignant lesions associated with *H. pylori*. COX-2 increases the level of Bcl-2 protein, which is involved in controlling apoptosis. An over-expression of COX-2 therefore leads to the development of resistance towards apoptosis, with dysregulation of cell death and cell growth in epithelial cells. This inhibition of apoptosis is associated with the initial phase of carcinogenesis (21).

There appears to be a strong association of environmental factors such as salt intake, nitrates and low intake of fresh fruits in the development of gastric cancer. *Fox et., al* (22) studied the effect of high-salt diet and *H. pylori* infection in transgenic hypergastrinemic mice and noted severe pathology with a combination of cell proliferation, pit cell hyperplasia and glandular atrophy. High salt switches Fas Ag-mediated apoptosis to proliferative signaling through MAPK-p38-mediated NF-B activation thereby, promoting neoplasia. Migrant populations when moving to areas of low risk show diminution of cancer rates

though this appears to be more gradual and depend on the age of the migration suggesting the role of environment as an important factor (23).

Conclusion

There is strong correlation between H.pylori and the incident of gastric carcinoma but no definite clue was found May be the only reliable evidence is the decline in the incident of gastric cancer after discovery infection.

Recommendations

The studies showed significant decline in the incidence of cancer after the discovery of the bacterium and introducing the eradication therapy. thus, programs and guidelines for treatment and eradication of the infection may actually become the key control of the gastric CA and also to decrease the incidence of peptic ulcers. educating people and the health workers about the bacterium and its complication and encouraging them to go to the health facilities for purposes of screening may indeed decrease the possible complications of *H. pylori* infection.

References

1. Roșu MC, Mihnea PD, Ardelean A, Moldovan SD, Popețiu RO, Totolici BD. Clinical significance of tumor necrosis factor-alpha and carcinoembryonic antigen in gastric cancer. Journal of medicine and life. 2022 Jan;15(1):4.
2. Hatakeyama M. Helicobacter pylori and gastric carcinogenesis. Journal of gastroenterology. 2009 Apr;44(4):239-48.
3. Uemura N, Okamoto S, Yamamoto S, Matsumura N, Yamaguchi S, Yamakido M, Taniyama K, Sasaki N, Schlemper RJ. Helicobacter pylori infection and the development of gastric cancer. New England journal of medicine. 2001 Sep 13;345(11):784-9.
4. Pandey R, Misra V, Misra SP, Dwivedi M, Kumar A, Tiwari BK. Helicobacter pylori and gastric cancer. Asian Pac J Cancer Prev. 2010 Jan 1;11(3):583-8.

5. Herrera, V. & Parsonnet, J. Helicobacter pylori and gastric adenocarcinoma. Clin. Microbiol Infect. 15, 971–976 (2009)
6. Muir CS, Harvey JC. Cancer of the stomach: Overview. In: Sugimura T, Sasako M, eds. Gastric cancer. London: Oxford University Press, 1997:3–21.
7. Bik EM, Eckburg PB, Gill SR, Nelson KE, Purdom EA, Francois F, Perez-Perez G, Blaser MJ, Relman DA. Molecular analysis of the bacterial microbiota in the human stomach. Proceedings of the National Academy of Sciences. 2006 Jan 17;103(3):732-7.
8. Moss SF. The clinical evidence linking Helicobacter pylori to gastric cancer. Cellular and molecular gastroenterology and hepatology. 2017 Mar 1;3(2):183-91.
9. Siurala M, Sipponen P, Kekki M. Campylobacter pylori in a sample of Finnish population: relations to morphology and functions of the gastric mucosa. Gut. 1988 Jul 1;29(7):909-15.
10. Forman D, Newell DG, Fullerton F, Yarnell JW, Stacey AR, Wald N, Sitas F. Association between infection with Helicobacter pylori and risk of gastric cancer: evidence from a prospective investigation. British Medical Journal. 1991 Jun 1;302(6788):1302-5.
11. Parsonnet J, Friedman GD, Vandersteen DP, Chang Y, Vogelman JH, Orentreich N, Sibley RK. Helicobacter pylori infection and the risk of gastric carcinoma. New England Journal of Medicine. 1991 Oct 17;325(16):1127-31.
12. Bass AJ, Thorsson V, Shmulevich I, Reynolds SM, Miller M, Bernard B, Hinoue T, Laird PW, Curtis C, Shen H, Weisenberger DJ. Comprehensive molecular characterization of gastric adenocarcinoma. Nature. 2014 Sep 11;513(7517):202.
13. Saadat I, Higashi H, Obuse C, Umeda M, Murata-Kamiya N, Saito Y, Lu H, Ohnishi N, Azuma T, Suzuki A, Ohno S. Helicobacter pylori CagA targets PAR1/MARK kinase to disrupt epithelial cell polarity. Nature. 2007 May;447(7142):330-3.
14. Bessede E, Staedel C, Acuna Amador LA, Nguyen PH, Chambonnier L, Hatakeyama M, Belleannee G, Megraud F, Varon C. Helicobacter pylori generates cells with cancer stem cell properties via epithelial–mesenchymal transition-like changes. Oncogene. 2014 Aug;33(32):4123-31.
15. Brandt S, Kwok T, Hartig R, König W, Backert S. NF-κB activation and potentiation of proinflammatory responses by the Helicobacter pylori CagA protein. Proceedings of the National Academy of Sciences. 2005 Jun 28;102(26):9300-5.

16. Wang F, Meng W, Wang B, Qiao L. Helicobacter pylori-induced gastric inflammation and gastric cancer. Cancer letters. 2014 Apr 10;345(2):196-202.
17. Hoffmann W. Stem cells, self-renewal and cancer of the gastric epithelium. Current medicinal chemistry. 2012 Dec 1;19(35):5975-83.
18. Ferrand J, Lehours P, Schmid-Alliana A, Mégraud F, Varon C. Helicobacter pylori infection of gastrointestinal epithelial cells in vitro induces mesenchymal stem cell migration through an NF-κB-dependent pathway. PLoS One. 2011 Dec 28;6(12):e29007.
19. Wei J, Nagy TA, Vilgelm A, Zaika E, Ogden SR, Romero–Gallo J, Piazuelo MB, Correa P, Washington MK, El–Rifai W, Peek RM. Regulation of p53 tumor suppressor by Helicobacter pylori in gastric epithelial cells. Gastroenterology. 2010 Oct 1;139(4):1333-43.
20. Cover TL, Blanke SR. Helicobacter pylori VacA, a paradigm for toxin multifunctionality. Nature Reviews Microbiology. 2005 Apr;3(4):320-32.
21. Nardone G, Rocco A, Vaira D, Staibano S, Budillon A, Tatangelo F, Sciulli MG, Perna F, Salvatore G, Di Benedetto M, De Rosa G. Expression of COX-2, mPGE-synthase1, MDR-1 (P-gp), and Bcl-xL: a molecular pathway of H pylori-related gastric carcinogenesis. The Journal of pathology. 2004 Mar;202(3):305-12.
22. Fox JG, Dangler CA, Taylor NS, King A, Koh TJ, Wang TC. High-salt diet induces gastric epithelial hyperplasia and parietal cell loss, and enhances Helicobacter pylori colonization in C57BL/6 mice. Cancer research. 1999 Oct 1;59(19):4823-8.
23. Peter S, Beglinger C. Helicobacter pylori and gastric cancer: the causal relationship. Digestion. 2007;75(1):25-35.
24. Bębnowska D, Grywalska E, Niedźwiedzka-Rystwej P, Sosnowska-Pasiarska B, Smok-Kalwat J, Pasiarski M, Góźdź S, Roliński J, Polkowski W. CAR-T cell therapy—an overview of targets in gastric cancer. Journal of Clinical Medicine. 2020 Jun;9(6):1894.