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PATHOGENESIS RISK FACTORS AND DIAGNOSIS OF *Helicobacter Pylori* INFECTION

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ABSTRACT

The spiral gastric bacteria colonizing acid secreting stomach are known to scientists for over 100 years. More recently, the discovery of *Helicobacter Pylori* and its association with peptic ulcer disease is confirmed. The mechanisms by which *H. pylori* cause mucosal inflammation and damage are not well defined but probably involve both bacterial and host factors. The persons with serological evidence of carrying cag-positive strains are at increased risk of developing both peptic ulcer disease and gastric carcinoma. Urease enzyme likely plays a significant role in the survival and growth of *H. pylori* in the stomach by creating an alkaline microenvironment. Poor hygiene, deficient sanitation and crowded conditions have been reported as risk factors. The diagnosis of *H.pylori* infection can be made either by invasively the endoscopy and biopsy or noninvasively serological analysis, urea breath test, urease detection or stool antigen tests. This paper reviews the pathogenesis, risk factors, and diagnosis of *H.pylori* infection.

KEY WORDS: *Helicobacter Pylori*, Pathogenesis, Risk Factors, Diagnosis

INTRODUCTION

Helicobacter Pylori discovered in 1983[1,2],bacterium that infects one half or more of World population[3]proved to have profound public–health implications[4,5].H.pyloricauses Chronicgastritis, peptic ulcer and gastriccancer[6,7]. Chronic gastritis and peptic ulcer diseases are common among the elderly and low-income groups [8,9]. Gastric cancer remains second among causes of cancer deaths worldwide. In the United States, annual direct costs of *H. pylori* associated disease exceed five billion dollar [10].There is a clear link between *H. pylori* infection and peptic ulcer. It has been estimated that 10 -20 % of *H.pylori* infected persons will develop a peptic ulcer in their life time[11].The relationship between *H.pylori* and gastric cancer was discovered.Gastric cancer of multistage degenerative process beginning with chronic gastritis[12].Studies in Europe, Latin America, China, and Japan aim to determine whether the *H. pylori* elimination prevents carcinoma or progression of precancerous lesions[13].The high frequency of gastric cancer in Asia led to their widespread screening program.Most studies examining the association between *H.pylori* gastric cancers have focused on older age patients when *H.pylori* prevalence is more common. Haruma et al. examined the frequency of *H.pylori* in patients with gastric cancer who were younger than 30 years of age [14].

Many studies favour*Helicobacter Pylori* infection in relation to other diseases, most notably heart disease [15, 16]but current evidence is inconclusive [17]. Helicobacter pyloriis detectable in nearly 95-100% of adult patients with duodenal ulcer and about 80% of patients with gastriccancer [18]. The suspected association between gastric ulcer and gastric cancer is now being confirmed [19].In developing countries where most children become infected by the age of 10,

gastric cancer rates is high.[20].Local Ministry of Health has confirmed an average of 80-85 deaths per annum due to gastric cancer from 2003-2005.

PATHOGENESIS

Helicobacter pyloriis able to survive in the gastric environments which are hostile to growth of other bacteria.When intraluminal (stomach) acidity diminishes due to gastric atrophy, H.pyloriis no longer able to colonize, possibly because of competing organisms.A major identified bacterial factors that permits gastric colonization include microaerophilism, for survival within the mucus gel; Spiral shape, and flagellate remain motile within the viscous layer; and urease production that generates ammonium that buffers the gastric acidity, and known to be toxic to cells.[21,22].Animals infected with H.pylori, indicate that both motility and urease activity are important determinants of ability to colonize the stomach[23].Although most organisms are free living in the mucus layer,smaller number appears to be adherent to the mucosal epithelial cells,forming "adherence pedestal"resembling those of entero pathogenicEscherichia coliH. pylorionlyoverly gastric-type but not the intestinal type epithelial cells. Affected gastric epithelial cells may be in the gastric ant rum or fundus or ectopic in the duodenum or in the esophagus[24-26].

Evidence that *H.pylori* causes pathological changes and is not just a secondary colonizer is now convincing[27].Primary evidence includes the development of gastritis after experimental ingestion in humans and by laboratory animals, and antimicrobial therapy that suppresses *H.pylori* normalizes tissue histology, but after relapse of infection the damage recurs[28,29].Supporting evidence includes the nearly universal humeral immune response, which is stable for years, but declines after with eradication of the organism[30,31], and the association of *H.pylori* with chronic diffuse superficial gastritis, but not other forms of gastritis (e.g.,Cohn'snon-steroidal anti-inflammatory drug (NSAID) - induced). Most peptic ulcers not caused by NSAID are now thought to be associated with Helicobacter. pylori infection[32].

The mechanism of tissue injury are not clearly established, and both bacterial and host factors may be determinants of outcome. *H. pylori* do not appear to invade tissues, as an incidental finding. Thus the lesions are likely to reflect a response to extracellular products of the organism. Ammonia,produced by urease and by deaminizes,is known to be toxic to eukaryotic cells,and may potentiate neutrophil induced mucosal injury. Gastritis is found in virtually all infected humans,although the majority have no symptoms; only I in 10 develop ulcer disease. Gastritis adenocarcinoma is 3 to 12 times more likely to develop in individuals infected with *H.pylori* [33,34].*H.pylori* is more likely to be associated with the early or initial states of primary gastric lymphoma than advanced tumours;*H.pylori* can disappear during progression of gastric lymphoma[35].About half of strains tested thus far produce vaculating cytotoxin (VacA), which induces acidic vacuoles in the cytoplasma of eukaryotic cells. 80-100 % patients with duodenal ulcer disease produce CagA antibodies against a 128 kd antigen compared with 60- 63 % of *H.pylori* infected persons-infected with gastritis only, indicating that serological responses to the 128 kd protein are more prevalent among H.pylori- infected persons with duodenal ulcer than infected persons without peptic ulceration[36,37].

Bacterial lipopolysaccharide usually has proinflammatory activities, but *H.pylori* lipopolysaccharide has little[38]. *H. pylori* lipopolysaccharide may express the type 11 Lewis(Lewis), neither of these antigens as well as type 1 antigens[39]. This observation is important because these antigens are present on gastric epithelial cells, and there is evidence that host Lewis phenotype selects for the particular Lewis expression of the *H. pylori* population[.40].The

presence of *H. pylori* overlying the gastric mucosa activates epithelial cells to produce proinflammatory cytokines and activates mononuclear and polymorph nuclear cells to produce cytokines, super peroxide, tumour necrosis factor-0, and other proinflammatory molecules[41].

Humans are polymorphic in the genetic loci involved in regulating proinflammatory cytokine production. Proinflammatory alleles regulating interleukin -1beta and interleukin-10 affect risk of gastric cancer, *H. pylori* positive persons. Virtually all patients with duodenal ulceration are colonized with cagA A(and thus cag Pathogenicity Island).Thus cagA, the first gene described not to be conserved among all *H.pylori* strains, is highly associated with both peptic ulcer disease and gastric cancer[42-44].Persons colonized with *H. pylori* have different gastric secretory physiology than do those who are not colonized. On average colonized persons have higher gastrin levels which are reduced by eradication of the organis[.45].

In recent years, it has become clear that stomach producehormones related to satiety and energy homeostasis, leptons and, to a greater extent, ghrelin, which has opposing effects. Studies indicate that H. pyloristatus affects the levels of these hormones and, in particular, *H. pylori* eradication leads to an increase in ghrelin. It is now clear that a generation of children in the developed countries is growing up *H. pylori* in their stomachs regulating these hormone[.46-48].In the last 10 years, there has been an increasing number of reports chiefly from East Asia, showing an epidemiologic association between the presence of *H. pylori* and the diagnosis and idiopathic thrombocytopenic purpura (IPT).Eradication of *H. pylori* has been attempted as a therapy for IPT, and although not conclusive, results are promising[49,50].

RISK FACTORS FOR *H.Pylori* INFECTION

Risk factors for *H. pylori* infection are now recognized. The prevalence of infection increases with age and low standard of living. The high prevalence reported in institutionalised patients and in submarine crews suggests that people living in close contact have high risk of infection [51-53].The same risk factors have been found been found for children as well as for adults, and high prevalence has also been reported in an orphanage[54,55].Recent work suggests that childhood could be the main period when *H. pylori* infection is acquired both in developing countries and in industrialised countries'[56].High risk of *H. pylori* infection in cohabiting persons has been confirmed by cases of familial coinfection, with evidence of the same strain among family members [57].Despite the generally high *H.pylori* prevalence throughout Asia, reported rates vary widely, Mitchell et al. found an overall prevalence of 44.2% in southern China, but there was significantly increased rate in urban areas (52.4 %) compared to 38.6 % in rural areas[58].General prevalence in nearby Taiwan was reported at 54.4 %[44].

Tibetan refugees in India had an overall prevalence of 77.2 %.Preliminary report of male Myanmar children (aged 5-14 years) living in monastery showed seroprevalence of 70.3 % with and inverse relation with age, and was not related to the duration of stay in the monaster[.59,60]. Seropravalence in the Vietnamese was 43 % in those 10-19 years of age but varied between 50 and 80 % in later years [61].Independent of socioeconomic status, ingestion of contaminated water is considered as likely mode of infection. Hopkins et al.noted, people who have uncooked vegetable consumption habits were more infected by H.pylorithan those who do not have them [62, 63].The absence of flush toilet in the house is associated with infection, supporting the hypothesis that the faecal-oral route may be important mechanism of transmission of this bacteriu[.64].In less than one century in United States, colonization has gone from being ubiquitous to being present in 5 % of children; this is a change in human micro ecology of major proportion[51].

DIAGNOSIS OF *H.Pylori* INFECTION

A. Invasive Methods Require Endoscopy

1. Culture

The demonstration of the organism by culture is the criterion of infection.Culturing for *H.pylori* involves obtaining the sample by endoscopy .Culture is no more sensitive than routine histological analysis. For these reasons,cultures are not indicated for diagnosis.[32]H.pylorigrowsin 3-6 days when incubated at 37C in a microaerophilic environments.The medium for primary isolation include Skirrow'smedium (Difco),with vancomycin, polymixin B,and trimethoprim,chocolate medium and other selective media with antibiotics (eg,vancomycin nalidixic acid,amphotericin). The colonies are non-haemolytic,mucoid,gray.

2. Endoscopy with Biopsy

1. Organism may have patchy distribution, especially in the body and fundus of stomach. Because the ant rum is more uniformly involved, two biopsy specimens from the prepyloric ant rum generally suffice.The yield may be increased further by sampling the fundus as well as the ant rum [32].
2. Routine haematoxylin and eosin staining may be unreliable for detecting *H. pylori* by microscopy. The Gymea and War thin-Starry stains permit easier visualization,especially by inexperienced observers[51].
3. Endoscopy is an expensive procedure,and several days may be required to obtain the stain/biopsy results.

3. Campylobacter like Organisms on Endoscopy Samples

Mucosal biopsy specimens may be directly inoculated into medium containing urea and phenol red-rapid urea test(RUT),which turns red if the pH rises above 6.0 .This change occurs when urea in the gel is metabolized to ammonia by the urease produced by H. pylori.This test is commercially available and inexpensive and can provide a diagnosis within 1 hour of inoculation of the biopsy specimen.Its sensitivity and specificity have been reported as high as 98 % and 100 % respectively, at 24 hours.The low cost and excellent reliability of this test make it the endoscopic method of choice for diagnosis [32].

B. Non-Invasive Tests

1**. Serological tests** are available, based on ELISAs.which have a reported sensitivity of 98 % and a specificity of 100 % for detecting IgG antibody to diagnose infection[32].Several ELISAs test kits are available eg,*H.pylori* One step test (Ultimed Pharma,Belgium),FlexiPack (Abbott,USA),Enzygnost Anti *H.pylori* 11 IgG (Siemens,Germany, *H.pylori* EIA KIT 96T (General Biological Corporation, Taiwan), and HEL-p-TestTM (Amrad,Australia).

1. An elevated antibody titter to *H.pylori* indicates current infection, because spontaneous clearance is rare.
2. There are several limitations to use of serology to document eradication of infection after therapy. Although the antibody titter falls after eradication,the rate of decline is uncertain.Titters must be following for at least 6 months to determine a decline. Whether a 20 or 50 % decline is IgG titter over 6 months suggests eradication is still undergoing evaluation.The excellent accuracy and low cost of serology make it the non-invasive method of choice to document infection with H.pylori; endoscopy is still required to diagnose ulcer disease [32].

2. **Urea breath test**have been developed because *H. pylori* has high urease activity.A solution containing urea labelled with carbon 13 or carbon 14 is ingested; after one hour, their breath is examined for carbon 13 or carbon 14,respectively. If *H.pylori* is present in the stomach, labelled carbon dioxide is split off by urease, absorbed and expired in the breath. Results of these assays correlate with numbers urease-producing *H.pylori* organisms and can be falsely negative after therapy that suppresses but does not eradicate the organism. However, negativity 1-3 months after therapy has ceased usually indicates eradication of infection [66].

Stool antigen tests relatively non-invasive. Most valuable for assessing response to eradication therapy after 6-8 week [.65].

CONCLUSIONS

H. pylori and its role in gastric cancer are confirmed. *H. pylori* cause inflammation in the stomach. Gastritis is found virtually in all infected humans. The mechanism by which *H. pylori* cause mucosal inflammation is not well defined. Low economics and poor living conditions are contributory factors. Diagnosis of *H. pylori* can be made by culture, endoscopy with biopsy, serological analysis and PCR.

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