INTRODUCTION

During the last 15 years, the role of Helicobacter Pylori (*H. pylori)* infection in the pathogenesis of gastritis and peptic ulcer disease in adults [(1)] and children [(2)] has been elucidated.

The Medicine Nobel Prize of 2005 was awarded to an observant pathologist Robin Warren and an enterprising physician Barry Marshal, both from Australia, for the discovery of Helicobacter pylori (*H. pylori*) and its role in peptic ulcer disease and gastritis in 1983[(3)]. *H. pylori* fulfill Koch's postulates because it is an infection causing chronic active gastritis: all infected persons have this histologically defined gastric inflammation, which in itself is asymptomatic but which may be associated with disease [(4)].

H. pylori show extensive genetic diversity, this variability has a crucial role in the pathogenesis of this bacterium [(5)]. Several *H. pylori* virulence factor genes related to severe clinical outcomes, including cytotoxin associated gene A (cagA), vacuolating cytotoxin gene A (vacA) and blood group antigen-binding adhesin gene (babA2), have been proposed[(5-8)]. Beyond the role of these factors in progression of the disease, there are several papers which reported a relationship between failure of *H. pylori* eradication therapy and the strains' virulence factor genotypes[(9,10)]. Therefore, *H. pylori* genotyping may have multiple roles including identification of the predominant *H. pylori* strains that are circulating in a given geographic area, impact on the cure rates of eradication therapy, and prediction of the clinical outcomes.[(6-10)]

The organism

Helicobacter pylori are a gram negative, curved, microaerophilic and motile organism with multiple polar flagella. They reside in the stomach of man and other primates, lining up the gastric mucous secreting cells[(11,12)]. Studies of the organism, biotyping, genome and plasmid profile diversity revealed the existence of approximately 40 epidemiological strains of *H. pylori* [(13)]. *H. pylori* is an obligatory microaerophilic bacteria, the optimal O2 concentration is 2.8%. In addition, CO2 is important for primary isolation. The basal media should be implanted with whole blood or serum and urea[(14)].

Epidemiology and geographical distribution:

H. pylori are one of the most common gastrointestinal bacterial infections among humans, affecting more than 50% of the world's population[(15,16)]. Infection is usually acquired during the first years of life in both developing and industrialized countries, with intra-familial spread playing a central role in transmission of the infection [(17,18)]. The prevalence of *H. pylori* is markedly variable between developing and developed countries, and even among individuals living in the same country, varying according to ethnicity, place of birth and socioeconomic factors. Besides geographic area, age is also significantly and independently associated with an increase in *H. pylori* prevalence, a phenomenon known as birth cohort effect, which is a progressive reduction of the infection rate in successive birth cohorts, due to the improvements in general living conditions [(19)]. In less developed countries the infection rate reaches almost 50% in very young children and more than 90% in adults, whereas in industrialized countries *H. pylori* infects 20-50% of adults and less

than 10% of children, and has been declining over time [19,20]. Indeed, the prevalence of *H. pylori* infection is showing a decreased trend worldwide that is directly associated with an improvement in the socioeconomic status and hygienic conditions of the populations[21].

Omran M (2003), reported positive titre of anti *H. pylori* IgA antibodies in 57.2% of the sample of Egyptian infants she studied[22]. Mohammed E (2004) detected a high prevalence of *H. pylori* infection (54.4%) among a group of healthy Egyptian children aged 3-12 years and the prevalence of *H. pylori* infection increased with age from 33.4% in those aged 3-7 years to 66.6% in children aged 8-12 years.

The overall prevalence of *H. pylori* infection in Egyptian schoolchildren was 72.38%. There was no significant difference in the prevalence of infection between boys and girls (73.80% vs. 70.34% respectively, odds ratio equal 1.05, P equal 0.63) [24].

Possible routes of transmission

Person-to-person transmission

Familial exposures.

Several studies performed DNA fingerprinting to determine the specific strains of *H. pylori* harbored by family members [25-28]. They found the same strain of *H. pylori* to be present in a small percentage of spouses and siblings[26-29]. These studies, taken as a whole, lend support to the concept of intrafamilial clustering of *H. pylori* infection. They suggest that person-to-person transmission occurred in these families possibly because of close interpersonal contact, that family members shared a genetic predisposition to *H. pylori* infection, that family members were exposed to a common source of infection, or that spouses' childhood socioeconomic class was similar.

Oral-oral route.

Many scientists have hypothesized that the oral-oral route of *H. pylori* transmission is the most likely, especially in developed countries. The most likely routes of oral infection include saliva, dental plaque, and refluxed gastric contents or vomit. Recent studies by several investigators have detected *H. pylori* DNA in saliva in H. pylori-positive subjects by using PCR assays [29-34].

fecal-oral route

Another possible method of *H. pylori* transmission is the fecal-oral route. *H. pylori* DNA has been detected in feces of infected subjects by some researchers [32,33,35] but not others [36]. However, isolation of *H. pylori* from feces has been problematic for some researchers, especially for those unable to obtain fresh feces. Delay in processing could have resulted in the small number of *H. pylori* organisms present being overgrown by other fecal bacteria [37].

Waterborne transmission

It has been demonstrated that *H. pylori* can live for several days in milk and tap water in its infectious bacillary form. A higher prevalence of *H. pylori* was found in the stools of

individuals who consumed well water and municipal tap water when compared to boiled water [38,39].

Zoonotic or vector borne transmission

Although the principal reservoir for *H. pylori* infection appears to be people, *H. pylori* have been isolated from non human primates and domestic cats [40-42].

Iatrogenic transmission

Because of the complex structure of the endoscope and difficulty in disinfecting it, the possibility of iatrogenic infection in patients following endoscopy is a potential risk factor not only for *H. pylori* but also for other infectious diseases such as hepatitis B, hepatitis C, tuberculosis, and possibly human immunodeficiency virus [43,44]. In fact, nosocomial transmission of *H. pylori* is the only proven mode of transmission[44].

Clinical picture of *H. pylori*

Children with *H. pylori* infection are often asymptomatic. Although non specific symptom have been associated with the presence of *H. pylori* gastritis in children, some clinical manifestation are reported to be more frequent [45-47].

Dyspepsia

The acute phase of colonization with *H. pylori* may be associated with transient nonspecific dyspeptic symptoms, such as fullness, nausea, and vomiting, and with considerable inflammation of both the proximal and distal stomach mucosa, or pangastritis. This phase is often associated with hypochlorhydria, which can last for months. It is unclear whether this initial colonization can be followed by spontaneous clearance and resolution of gastritis and, if so, how often this occurs[48]. Follow-up studies of young children with serology or breath tests suggested that infection may spontaneously disappear in some patients in this age group[49-51]; this has not been observed in adults other than under specific circumstances, such as development of atrophic gastritis[48].

Non-ulcer dyspepsia is defined as the presence of dyspeptic symptoms in the absence of endoscopic findings[52]. An increased prevalence of *H. pylori* has been reported in this condition, but inconsistent long term symptom relief has been observed with bacterial eradication in large, randomized trials[53,54]. The reason for these discrepant results is not entirely clear. A Coch-rane review suggests that eradication of *H. pylori* improves symptoms in only about 9 percent of patient with dyspepsia without ulcers, but this end point may miss the other potential benefits of *H. pylori* eradication[55].

Dyspeptic symptoms may have a reflux-like character, with heart pain and regurgitation as predominant signs or may appear as dysmotility-like, with early satiety and nausea or may be ulcer-like pain and vomiting. Thirty to 60% of patients with functional dyspepsia carry *H. pylori*, but this prevalence is not much different from that in the unaffected population[56]. *H. pylori* play a role in the etiology of dyspeptic symptoms; thus *H. pylori* test-and-treat strategies are effective for this subgroup of patients with dyspepsia[57].

Gastroesophageal reflux

Eradication has become an issue in patients with gastroesophageal reflux disease, since long –term acid suppressive therapy may aggravate *H. pylori*-mediated corpus gastritis and increase the risk of gastric carcinoma[58].

Controversially, some case-control and cohort studies have suggested that *H. pylori* infection may protect against gastroesophageal reflux disease. Two recent, fully controlled trials showed, however, that *H. pylori* eradication did not negatively influence relapse rates in patients with gastroesophageal reflux disease[59,60], but additional prospective studies are needed.

Other cross sectional observations suggested that the prevalence of *H. pylori* among GERD patients was similar to that among controls[61].

Gastrointestinal bleeding

Another symptom related to *H. pylori* infection in children is gastrointestinal bleeding (hematemesis or melena)[62]. This relation was suggested by many studies[63,64]. Shalaby S (2004) confirmed the presence of *H. pylori* in 35%of 37 Egyptian children aged one to twelve years, presenting with hematemesis by upper gastrointestinal endoscopy, histological examination and rapid urease test (RUT) of biopsied specimens[65].

Even microscopic bleeding that may be sufficient to cause iron deficiency anemia was found to occur in children with active *H. pylori* infection. Blecker et al (1991) reported severe iron deficiency anemia in a child with *H. pylori* associated chronic superficial gastritis with no gastrointestinal symptoms, the anemia subsided after *H. pylori* eradication[61].

Chronic or recurrent abdominal pain:

The definition of chronic abdominal that was used clinically and in research over the last 40 years is based on the criterion of at least 3 pain episodes occurring over at least 3 months and interfering with normal functions. In clinical practice, it is generally believed that pain which exceeds 1 or 2 months in duration can be considered chronic[67]. Chronic abdominal pain which affects normal activity or usual levels of performance has been reported to occur in 10-15% of children and adolescents between the ages of 4-16 years[62].

Pain is poorly localized and usually periumblical: it is often severe but short- lived, rarely interferes with appetite or voluntary activity, rarely awaken the child, and may occur at specific times (for example, on school mornings). There are some clues suggesting an organic etiology for RAP, such as night- time pain, oral regurgitation, recurrent vomiting, heartburn, epigastric pain, growth failure, and failure to respond to standard approaches. Some authors consider RAP and dyspepsia as different entities, based on localization and associated symptoms[68,69].

Chronic or recurrent abdominal pain is considered the main presentation of *H. pylori* infection in children in many studies[63,70-73]. In the study of De Giacomo et al (1992), the authors concluded also that treatment of *H. pylori* infection in those children resulted in amelioration of symptoms[70]. However, other investigators disputed this relation as their results did not

demonstrate evidence for the association of *H. pylori* infection and RAP in children[74-76].Thus the role of *H. pylori* infection in children with RAP remains highly contentious.

The effect on nutrition and growth

Studies from Italy, Germany and USA have shown that H pylori infection is associated with growth delay especially in older children.[77,78] However, it is not yet clear whether the difference in anthropometry between *H. pylori* infected and non-infected children is solely due to *H. pylori* infection or the socioeconomic and ethnic factors also contribute to it. We need more information, especially from developing countries where *H. pylori* infection is rampant in children, before accepting that *H. pylori* cause growth retardation.

Gharlin, produced in stomach, is a hormone that stimulate food intake and weight gain, but controversy exists over the relationship between ghrelin release and *H. pylori* infection. Plasma levels of circulating ghrelin decrease during *H. pylori* infection and following *H. pylori* eradication, ghrelin levels increase significantly.[79]

Decrease ghrelin concentration are also associated with higher levels of leptin[79], a hormone responsponsible for suppressing appetite, regulating energy intake and modulating energy expenditure, and a reduction in the secretion of growth hormone[80]. There is some evidence therefore that the negative association between ghrelin and *H. pylori* infection may be contribute to malnutrition and growth retardation.

Iron Deficiency Anemia and *H. pylori* Infection

There is some suggestion that *H. pylori* causes iron deficiency anemia (IDA) especially in adolescent girls without producing any hemorrhagic lesions in the stomach or duodenum. Kostaki, et al.[81] from Greece first time reported that IDA in 3 children improved only after *H. pylori* eradication. Subsequently a report from Korea[82] on 937 children has shown that *H. pylori* infection was more common in children with IDA (35.5%) than in children without IDA (19.4%). A recent report from Turkey[83] on 140 children (6 to 16 years) has shown that iron deficiency (ID) and iron deficiency anemia (IDA) improved completely after *H. pylori* eradication without any iron supplementation. The postulated mechanisms for IDA in *H. pylori* infection are: poor absorption of iron due to low gastric acid secretion, poor dietary intake and consumption of iron by the bacteria itself.

Other disorders:

These include coronary heart disease, dermatological disorders such as rosacea and idiopathic urticaria, autoimmune thyroid disease, thrombocytopenic purpura, Reynaud's phenomena, scleroderma, migraine, and Guillian-Barre syndrome. The underlying hypothetical mechanisms include chronic low- grade activation of the coagulation cascade, accelerating atherosclerosis, and antigenic mimicry between *H. pylori* and host epitopes leading to autoimmune disorders[84].

Pathobiology:

Only a small percentage of children or adults infected with *H. pylori* will progress to chronic active gastritis, peptic ulcer disease, and/ or gastric cancer. Disease progression depends on interplay, between bacterial factors, host genetic background and environmental factors. Identifying bacterial markers to distinguish those at risk of peptic ulcer or gastric cancer from those not at risk would be significant advance in the management of *H. pylori* gastritis particularly in children[85]. The gastric mucosa is well protected against bacterial infections. However, *H. pylori* is highly adapted to this ecologic niche, with a unique array of features that permit entry into the mucus, swimming and spatial orientation in the mucus, attachment to epithelial cells, evasion of the immune response, and as a result, persistent colonization and transmission several factors contribute to its virulence.

The *H. pylori* genome (1.65 million bp) codes for about 1500 proteins[86,87]. Among the most remarkable findings of two *H. pylori* genome –sequencing projects were the discovery of a large family of 32 related outer-membrane proteins (OMP proteins) that includes most known *H. pylori* adhesins and the discovery of many genes that can be switched on and off by slipped-strand mispairing-mediated mutagenesis. Proteins encoded by such phase-variable genes include enzymes that modify the antigenic structure of surface molecules and control the entry of foreign DNA into the bacteria. The genome of *H. pylori* changes continuously during chronic colonization of an individual host by importing small pieces of foreign DNA from other *H. pylori* strains during persistent or transient mixed infections[88,89].

After being ingested, the bacteria have to evade the bactericidal activity of the gastric luminal contents and enter the mucous layer. Urease production and motility are essential for this first step of infection. Urease hydrolyses urea into carbon dioxide and ammonia, thereby permitting *H. pylori* to survive in an acidic milieu[90]. Several reports demonstrated the toxic effect of ammonia on the gastric mucosa as it impairs mitochondrial and cell respiration thus decrease cell viability[91].

Other bacterial enzymes that affect the mucous layer are mucinase, lipase and phospholipase; they all degrade the mucous by their activity on proteins and lipids. Catalase is another virulence factor which protects the bacteria against active oxygen radicals produced by the neotrophils[92]. Motility is essential for colonization, and *H. pylori* flagella have adapted to the gastric niche[93].

H. pylori can bind tightly to epithelial cells by multiple bacterial-surface components[94]. The best-characterized adhesin, blood group antigen-binding adhesin gene (babA), is a 78-kD outer-membrane protein that binds to the fucosylated Lewis B blood-group antigen [95]. Studies with transgenic mice expressing the human Lewis b epitope in gastric epithelial cells indicated that Lewis b functions as a receptor to gastric pit and surface mucous cells[96]. Attachment of *H. pylori* to gastric epithelial cells in such transgenic mice resulted in the development of chronic gastritis and gastric atrophy[97]. Recently, the gene encoding babA has been cloned (and termed babA2), which thus allows identification of *H. pylori* strains harboring the babA2 genotype by PCR[95].

The majority of *H. pylori* strains express the 95-kD vacuolating cytotoxin gene A (vacA), a secreted exotoxin[98]. The mature toxin is peculiarly suited to the gastric environment in

that it is activated by acid (even if the pH is subsequently neutralized) and in its activated form it not only causes more profound epithelial changes than before activation, but also becomes resistant to subsequent damage by acid and pepsin[99]. The toxin inserts itself into the epithelial-cell membrane and forms a hexameric anion-selective, voltage-dependant channel through which bicarbonate and organic anions can be released[100], possibly providing the bacterium with nutrients.

VacA is also targeted to the mitochondrial membrane, where it causes release of cytochrome c and induces apoptosis[101]. The pathogenic role of the toxin is still debated. Although only about 40% of *H. pylori* are toxigenic, all have vacA, the gene encoding the toxin. VacA-negative mutants can colonize in animal models, and strains with inactive vacA genes have been isolated from patients, indicating that vacA is not essential for colonization. However, vacA-negative mutants were outcompeted by wild-type bacteria in a mouse model, indicating that vacA increase bacterial fitness in this model[102].

Significant sequence polymorphism within vacA can be found in the coding sequence for the signal peptide (referred to as the s-region) and in the middle of the gene, called the middle (m) region. There are two allelic types (m1 and m2) in the middle region while the signal region has either an s1 (s1a, s1b, and s1c) or an s2 allele.

The strains of the s1/m1 subtype typically produced higher levels of the vaculating cytotoxin than other genotypes, while s2/m2 strains do not secrete[103]. Strains with vacA s1a alleles were most commonly isolated from patients with ulcers (89% of patients with s1a strains had past or present ulcers) whereas strains with vacA s2 alleles were uncommonly isolated from ulcer patients (20% of those with s2 strains had ulcers)[104].

Toxins with different m- genotypes also display a differential for intoxicating the target mammalian cells, with vacA m1 variants affecting wider range of target cells than those with m2[105].

Approximately 50% to 60% of *H. pylori* contain the cagA gene, encoding the cagA protein[106]. The name cagA (cytotoxin associated gene A) is misleading as disruption of cagA does not affect production or activity of vaculating cytotoxin[107]. The cagA gene is part of cag Pathogenicity Island (cag PAI), which contains many genes that are related to the virulence and pathogenicity of *H. pylori* strain[108]. The presence of cagA is a confirmed marker for cag PAI and is associated with more virulent *H. pylori* strains[109].

The product of the cagA gene is introduced into gastric epithelial cells by the type IV secretion system, where it becomes phosphorylated and modulates various cellular process and signal transduction pathways. The intracellular cagA activities associated with the development of gastric carcinoma include disruption of tight junctions[110]. The presence of cagA gene is an important marker for the most virulent strains associated with peptic ulcer, atrophic gastritis and adenocarcinoma[111].

Pathology

The pattern and distribution of gastritis correlate strongly with the risk of clinical sequel, namely duodenal or gastric ulcers, mucosal atrophy, gastric carcinoma, or gastric lymphoma[112]. Patients with antral-predominant gastritis, the most common form of *H. pylori* gastritis, are predisposed to duodenal ulcers, whereas patients with corpus-predominant gastritis and multifocal atrophy are more likely to have gastric ulcers, gastric atrophy, intestinal metaplasia, and ultimately gastric carcinoma[113].

The natural history of *H. pylori* infection can be thought of in two phases (figure 1)[114]

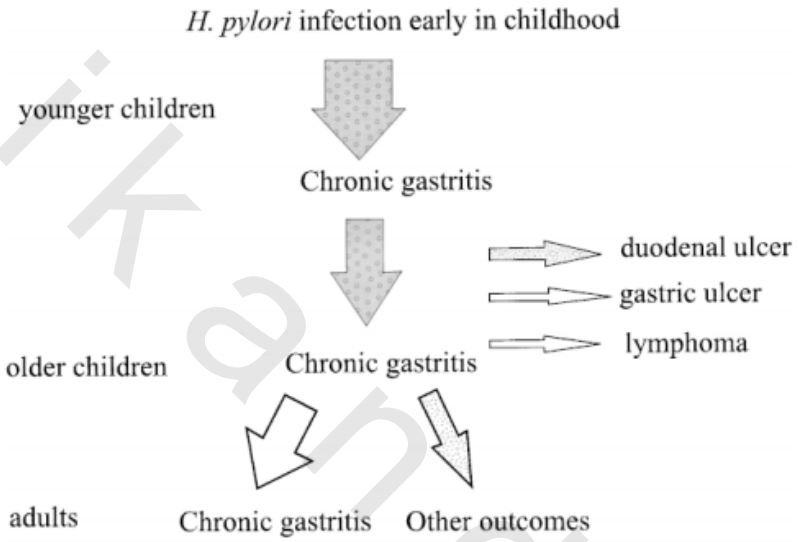

Figure (1): The natural history of *H. pylori* infection can be thought of in two phases *H. pylori* infection in children is associated with a number of potential outcomes. However, most of the children will develop chronic superficial gastritis without any symptomatology. A minor group of children will develop duodenal ulcer as older children or adolescents. MALT lymphoma and gastric ulcers are very rarely seen in pediatrics.

Acute gastritis

Is defined as microscopic evidence of inflammation affecting the gastric mucosa. The intensity of the inflammatory response is variable. Mild mucosal inflammation may be difficult to distinguish from normal mucosa and often requires review by an experienced pathologist[115].

Moreover, the acute phase of helicobacter infection is rarely encountered in gastric biopsy specimens because the initial illness has trivial symptomatology and goes unnoticed by the patient[50].

Chronic inflammation

H. pylori consider the commonest cause of chronic active gastritis[116]. It is generally considered to be present if there are more than 2-5 lymphocytes, plasma cells and macrophages per high power field[117]. In children with *H. pylori* infection, substantial

numbers of plasma cells and lymphocytes are present in the mucosal biopsy sections. The inflammatory cellular infiltrate is usually superficial in location, with panmucosal inflammation present in a small number of cases. The term activity is used to characterize the presence of neotrophil in the gastric biopsy. However, neotrophil activity is almost always present in *H. pylori* infection in adults more than in children[118].

When colonization does become persistent, a close correlation exists between the level of acid secretion and the distribution of gastritis. This correlation results from the counteractive effects of acid on bacterial growth versus those of bacterial growth and associated mucosal inflammation on acid secretion and regulation. This interaction is crucial in the determination of outcomes of *H. pylori* infection.

In subjects with intact acid secretion, *H. pylori* in particular colonizes the gastric antrum, where few acid-secretory parietal cells are present, in these subjects colonization pattern is associated with an antrum-predominant gastritis, histological evaluation of gastric corpus specimens in these cases reveals limited chronic inactive inflammation and low numbers of superficially colonizing *H. pylori* bacteria.

Subjects in whom acid secretion is impaired, due to whatever mechanism, have a more even distribution of bacteria in antrum and corpus, and bacteria in the corpus are in closer contact with the mucosa, leading to a corpus-predominant pangastritis[119].

The reduction in acid secretion can be due to a loss of parietal cells as a result of atrophic gastritis, but it can also occur when acid secretory capacity is intact but parietal cell function is inhibited by vagotomy or acid-suppressive drugs, in particular, proton pump inhibitors[119].

The resulting active inflammation of the corpus mucosa further augments hypochlorhydria, paralleling the acute phase of infection, as local inflammatory factors such as cytokines, including interleukin-1B (IL-B), have a strong suppressive effect on parietal cell function. This is illustrated by various observations, firstly, *H. pylori* corpus gastritis is often associated with hypochlorhydria, and eradication therapy leads to increased acid secretion in these subjects [61,120] , secondly, *H. pylori* corpus gastritis augments the acid-suppressive effects of PPIs[121].

As a result, *H. pylori* positive patients with gastro-esophageal reflux disease (GERD) may respond somewhat faster to PPI treatment both with respect to symptom resolution and with healing of esophagitis[122], but this effect is minimal and largely irrelevant in daily clinical practice. This means that there is no general need to take *H. pylori* status into account when decisions on the dose of PPI treatment for GERD must be made.

A third observation in support of the acid-suppressive effects of active corpus gastritis comes from more recent, important research showing that subjects with proinflammatory genotypes have a higher risk of corpus-predominant pangastritis, predisposing them to atrophic gastritis, intestinal metaplasia, and gastric cancer[123].

Although gastric colonization with *H. pylori* induces histologic gastritis in all infected individuals, only a minority develop any apparent clinical signs of this colonization. It is estimated that *H. pylori*-positive patients have a 10 to 20% lifetime risk of developing ulcer disease and a 1 to 2% risk of developing distal gastric cancer[124-126].

Duodenitis is characterized by the presence of neotrophil in the lamina propria, crypts or surface epithelium, in addition to an increase in the number of mononuclear cells.

Peptic ulcer is a deep mucosal lesion that disrupts the muscularis mucosa coat of the gastric or duodenal wall, while peptic erosions are superficial mucosal leisions that do not penetrate the muscularis mucosa [127].

Peptic ulcer disease:

In initial reports from all over the world in the first decade after the discovery of *H. pylori*, approximately 95% of duodenal ulcers and 85% of gastric ulcers occurred in the presence of *H. pylori* infection [126]. Several cohort studies estimated that lifetime risk for ulcer disease in *H. pylori*-positive subjects is 3 to 10 times higher than in *H. pylori*- negative subjects[128] and that 10 to 15% of *H. pylori*-positive subjects developed ulcer disease during long-term follow-up[129].

Ulcers mostly occur at sites where mucosal inflammation is most severe[130]. In subjects with decreased acid output, this usually is the gastric transitional zone between corpus and antrum, giving rise to gastric ulcer disease. If acid production is normal to high, the most severe inflammation usually is found in the distal stomach and proximal duodenum, giving rise to juxtapyloric and duodenal ulcer disease[131,132].

Almost all peptic ulcers in children are located in the duodenum, whereas gastric ulcers are extremely rare in this population[133]. Similar to the findings in adults, the occurrence of duodenal ulcers in the absence of *H. pylori* is uncommon in children unless they are being administrated nonsteroidal anti-inflammatory drugs[134,135]. Moreover, it has been demonstrated that duodenal ulcer disease in children does not relapse if infection with *H. pylori* is cleared from gastric mucosa[133].

Endoscopic features

Endoscopy is the only method to accurately diagnose peptic ulceration in children[136,137]. A nodular mucosa in the gastric antrum or duodenal bulb and/or gastric or duodenal erosions or ulcerations are specific (but not sensitive) features, suggesting active *H. pylori* infection. For those with suspected infection, biopsies should be obtained for histopathology, as well as complementary tests for detection of *H. pylori* including rapid urease test, histopathology with Giemsa stain and, if available, culture. The rationale for the recommendation to perform more than one diagnostic test is based on their sensitivity results in children, which range from 66% to 100% for histology and from 75% to 100% for rapid urease tests[138].

Histology

Histology can provide information about tissue morphology (e.g. gastritis or nodular gastritis)[139,140] and this is highly sensitive and specific for *H. pylori* infection and also has the advantages that specimens can be reexamined. Although there is no specific stain for, *H. pylori* two staining method are commonly used. The whartin-starry silver stain is the most preferable technique; Giemsa stain is alternative considered by some investigators to be equivalent in quality to silver stain[141]. Because histologic gastritis may be present in the absence of macroscopic mucosal abnormalities, this examination permits a better correlation between the presence of *H. pylori* and its pathologic results.

As *H. pylori* reside exclusively in the gastric mucosa with the highest concentration of the bacteria in the antral area, therefore, biopsies from the antrum are the preferred site for histological examination.

The majority of children with *H. pylori* infection have moderate to severe antral gastritis characterized by infiltration of lymphocyte and plasma calls in the lamina propria, and in severe cases foci of active inflammation within the glandular epithelium are also seen. In some cases, *H. pylori* organisms can be recognized in the sections, their characteristic appearance is 3.0x0.5 um spiral rods, located at the gastric mucosa adjacent to the gastric epithelium[141,142].

Lymphoid follicles with germinal centers are usually seen particularly in the deeper portion of the mucosa. This finding, which is easily to identify on low-power microscopic examination of biopsies, is virtually pathognomonic for the presence of H. pylori[143].

The surface and pit- lining epithelium is infiltrated by neutrophils, which may be so prominent that pit abscesses are formed. This neutrophil infiltration is termed "active gastritis" and is seen predominantly in areas where the helicobacter organisms are most abundant and most readily identified[144].

Reactive epithelial changes accompany active gastritis and consist of nuclear enlargement and prominent nucleoli. On occasion, these reactive changes may be so prominent that difficulty is experienced in distinguishing them from dysplasia. The problem is similar to that encountered in distinguishing reactive changes in ulcerative colitis from colonic dysplasia[145].

The single most useful criterion for the identification of inflammatory atypia is the presence of an associated neutrophillic infiltrate.

It is therefore, recommended that in acutely inflammed mucosa, a positive diagnosis of dysplasia should be made with the extreme reluctance. It is better to issue a noncommittal report and request that repeated biopsies be taken after an interval of antibiotic therapy[144].

Treatment of *H. pylori* gastritis with appropriate antibiotic will usually result in rapid elimination of organism and abolition of neotrophilic infiltrate. Chronic inflammation however, may persist for several years before disappearing[146,147].

Multifocal Atrophic Gastritis

Multifocal atrophic gastritis (MAG) affects both the pyloric and fundic mucosa in a patchy fashion. The characteristic histological feature is atrophy of the glands, which is accompanied by varying degrees of full-thickness chronic inflammation.

Intestinal metaplasia is invariably present in the later stages but may not be found with minor degrees of atrophy.

Early atrophy may be difficult to recognize in situations in which there is an intense full-thickness infiltrate in the lamina propria that pushes apart adjacent glands[144].

Because minor degrees of atrophy do not seem to be clinically important, it is recommended therefore that MAG should be diagnosed only when there is unequivocal

evidence of mucosal atrophy. For practical purposes, this means the presence of intestinal metaplasia, intestinal metaplasia is typically present as small patches of goblet cells and Paneth cells without the formation of villi. Adjacent to these patches, the mucosa may show active inflammation with pit abscesses formation[148].

However as intestinal metaplasia progress, the gastric microenvironment changes, so that inflammation lessens and *H. pylori* organisms are eliminated[149]. MAG is the commonest pattern of chronic gastritis in the third world[150] whereas DAG is the commonest pattern in westernized countries.

The generally held opinion now is that most, but not necessarily all, examples of multifocal atrophic gastritis represent the end of *H. pylori* gastritis in which the organism have damaged the mucosa so severely that the intragastric environment is altered and condition become inhibitory to further bacterial growth[149,151,152]. Longitudinal studies suggest that, in all patients with gastritis the severity of the disease and the extent of atrophy increase with advancing age[153]. However this does not exclude the possibility that multifocal atrophic gastritis has other causes.

Factors promoting atrophy and metaplasia in *H. pylori* gastritis are ill understood.

However there are theoretic grounds for believing that:

1- A lack of dietary vitamin C.[154,155]
2- Intragastric nitrosamine formation.[155]
3- Salt consumption.[156]
4- Auto antibodies that cross-react with gastric antigens demonstrated in patients with *H. pylori* -induced atrophic gastritis[157]. It appears possible therefore, that autoimmunity may play a role in disease progression

Because of epidemiologic and prognostic difference, it now seems reasonable to identify separately non atrophic and atrophic forms of *H. pylori* gastritis as DAG and MAG respectively. In advanced MAG, there may be hypochlorhydria, but in contrast to autoimmune gastritis, achlorhydria and pernicious anemia do not occur[144].

Sydney system is a novel classification and grading of gastritis that was devised by a group of experts at the 9th World Congress of Gastroenterology in Sydney, Australia in 1990. In 1994 in Houston, Texas, experts devised the new updated Sydney system[48,158,159].

Sydney System had both endoscopic and histologic divisions. The histologic arm emphasized the importance of combining topographical, morphological, and etiological information into a scheme that would help generate reproducible and clinically useful diagnoses[117,160].

In Sydney System grading of gastritis new visual analog scale for the graded variables and a set of guidelines for its application have been designed[116].

Different genotypes of *H. pylori* circulate in different geographic areas; the different clinical presentations are partly attributed to this high genetic diversity; however no clinical study has focused on the clinical and endoscopic findings of different genotypes so far. In this study, we aimed to investigate the probable relationship between virulence

factors and clinical, endoscopic, and histopathological findings of children with *H. pylori* infection.