Abstract
Helicobacter pylori represents one of the most common and medically prominent infections worldwide. Infection with this microaerobic, gram-negative bacterium has been established as an etiologic factor in the development of peptic ulcer disease. In addition, H pylori infection has been associated firmly with the development of gastric neoplasia, including gastric adenocarcinomas and gastric mucosa-associated lymphoid tissue lymphomas. Effective antimicrobial treatment depends on sensitive and accurate diagnostic approaches. This review article discusses invasive and noninvasive strategies for diagnosis of H pylori infection. Invasive methods requiring endoscopic evaluation include bacteriologic culture and susceptibility testing, histopathologic studies, molecular diagnostics, and rapid urease testing. Noninvasive approaches include fecal antigen detection, serologic testing, and urea breath testing.

In 1983, Warren and Marshall proposed the possible association of Helicobacter pylori with peptic ulcer disease and gastric cancer. In February 1994, the National Institutes of Health Consensus Development Conference concluded that H pylori infection represents the major cause of peptic ulcer disease, and all patients with documented peptic ulcer associated with H pylori infection should receive antimicrobial therapy. The risks of ulcer recurrence and associated complications do not diminish unless H pylori infection is cured. In June 1994, the International Agency for Research on Cancer Working Group of the World Health Organization classified H pylori as a group I, or definite, human carcinogen. Highlighting its significance in global infectious diseases, H pylori has been an important target in bacterial genomics as the first bacterial pathogen with entire genomes sequenced from 2 different strains.

The prevalence of peptic ulcer disease exceeds 6.5 million cases in the United States, approximating a rate of 2.5%, or 2,500 cases per 100,000 individuals. In developed countries, the overall prevalence of H pylori infection ranges from 25% to 30%. Seroprevalence increases with age, ranging from 5% to 27% in early childhood to levels exceeding 50% in adults older than 50 years. Seroprevalence studies demonstrate an acquisition rate in adults of 3% to 4% per decade. H pylori infection is a likely contributing factor in the development of gastric neoplastic diseases such as gastric adenocarcinoma and mucosa-associated lymphoid tissue (MALT) lymphoma. H pylori infection is considered to increase the risk of gastric adenocarcinoma with pooled odds ratios of 1.9 to 2.5.

H pylori and its association with multiple gastroduodenal diseases have emphasized the importance of accurate and
prompt diagnosis of symptomatic individuals. The “test and treat” approach may be justified on the basis of a significantly increased lifetime risk of chronic peptic ulcer disease and gastric cancer. In this review, the basic features of \( H \) pylori infection and diagnostic testing are described. Excellent reviews of \( H \) pylori infection and antimicrobial therapy, gastric adenocarcinoma, MALT lymphoma, pathogenesis, and diagnostic testing strategies have been published. Recent advances in basic molecular biology and treatment of \( H \) pylori will not be explored further in this review.

**Gastro-duodenal Pathology**

Immediately following infection, \( H \) pylori causes acute gastritis characterized by neutrophil infiltration into the foveolar and surface epithelium and epithelial degenerative changes. \( H \) pylori causes a persistent infection in the majority of infected individuals. The acute phase lasts 1 to 4 weeks and is replaced gradually by a chronic, mononuclear infiltrate in the lamina propria. Active gastritis refers to the presence of neutrophils mixed with mononuclear cells in the gastric mucosa. Chronic active gastritis occurs in the majority of infected individuals and consists of surface epithelial degeneration, persistent neutrophil infiltration of the epithelium and lamina propria, and mononuclear infiltration (lymphocytes and plasma cells) of the lamina propria. Lymphoid hyperplasia in the gastric mucosa is suggestive of \( H \) pylori infection. Generally, gastritis is most prominent in the corpus and antrum, with evidence of inflammation of the cardia in most infected individuals.

Long-term infection by \( H \) pylori results in chronic gastritis, a condition manifest as multiple pathologic entities (Figure 1). Chronic gastritis due to \( H \) pylori infection may be separated into distinct, clinically relevant phenotypes. Nonatrophic gastritis occurs in the majority of \( H \) pylori–infected individuals with no predisposition to peptic ulcer disease or gastric atrophy. Prominent mucosal inflammation in chronic active gastritis often is evident in the antrum (antral-predominant gastritis), predisposing to hyperacidity and duodenal ulcer disease. In contrast, multifocal atrophic gastritis and atrophic corpus-predominant gastritis result from long-standing infection and are characterized by glandular atrophy, intestinal metaplasia, and sparse inflammatory cells. Both forms of atrophic gastritis and the presence of intestinal metaplasia are associated with an increased risk of gastric adenocarcinomas. In addition, lymphocytic and granulomatous gastritis have been linked with \( H \) pylori infection. Although isolated cases of idiopathic granulomatous gastritis have been demonstrated in association with \( H \) pylori infection, it is unclear whether \( H \) pylori has an important role in the development of gastric granulomas. The majority of cases of granulomatous gastritis have been linked with Crohn disease, foreign body reactions, and sarcoidosis. In contrast with granulomatous gastritis, \( H \) pylori infection seems to have an important role in the pathogenesis of lymphocytic colitis. In a subset of cases associated with \( H \) pylori infection, specific antimicrobial treatment and eradication of \( H \) pylori infection have been linked with histologic improvement, including a decline in the number of intraepithelial lymphocytes.

Bacterial and host factors seem to influence the type of gastroduodenal disease that is generated. As many as 16% of \( H \) pylori–infected individuals in the United States develop duodenal ulcers in addition to chronic active gastritis. An inverse relationship or paradox exists between the incidence of duodenal ulcer disease and gastric adenocarcinoma. Patients with duodenal ulcers rarely develop gastric adenocarcinoma and vice versa. In a recent study, no patients with duodenal ulcers developed gastric adenocarcinoma, whereas 3.4% of patients with gastric ulcers developed gastric adenocarcinoma. In addition, 4.7% of patients with nonulcer dyspepsia and 2.2% of patients with hyperplastic polyps developed gastric adenocarcinoma. Patients with antral-predominant gastritis are at increased risk for duodenal ulcer disease. In contrast, multifocal atrophic gastritis is a common precursor state for both gastric ulcers and gastric adenocarcinomas and is not associated with duodenal ulcer disease. Individuals infected in early childhood are especially at risk for the development of multifocal atrophic gastritis and subsequent gastric adenocarcinoma. Atrophic changes in the gastric mucosa and the presence of intestinal metaplasia have been linked to the development of gastric adenocarcinoma. Elevated rates of early childhood infection in developing countries may explain, at least in part, the greatly increased prevalence of gastric adenocarcinoma in these countries.
locations. Seroepidemiologic studies indicate that infected individuals are at 6-fold increased risk for gastric MALT lymphoma. If left untreated, a subset of gastric marginal zone B-cell lymphomas (low-grade gastric MALT lymphomas) may give rise to diffuse large B-cell lymphomas (high-grade MALT lymphomas) that are refractory to therapy.

**Histopathologic Evaluation of *H pylori* Infection**

Endoscopically, gastric infection with *H pylori* is characterized by glands that are atrophic and associated with lymphocytic infiltrates, mononuclear cells, a balanitis pseudodiffuse lymphoid infiltrate, and intestinal metaplasia. 

Esophagogastroduodenal (EGD) endoscopy permits gross visualization and localization of ulcerative lesions, mucosal nodularity associated with MALT lymphomas, and other malignant lesions. Infection with *H pylori* can be patchy, and, as a result, multiple biopsy specimens may be necessary for diagnosis. Two gastric mucosal biopsy specimens (from antrum and corpus) usually suffice for the histologic diagnosis of *H pylori* infection and *H pylori*-induced gastritis. If 2 or more biopsy specimens are obtained, the sensitivity and specificity of histologic studies for the diagnosis of *H pylori* infection exceed 95%. In contrast, sampling of single sites may result in diminished sensitivities.

In patients with suspected *H pylori*-associated gastritis or peptic ulcer disease, antral inflammation and the presence of *H pylori* organisms are evaluated in mucosal biopsy specimens. More than 90% of duodenal ulcers are associated with *H pylori*, which is present in highest concentrations in the gastric antrum. A proximal-distal gradient of increasing organism densities exists along the corpus and antrum in duodenal ulcer disease and extends toward the transitional zone and gastroduodenal junction. Consequently, virtually all patients with duodenal ulcer disease have chronic, active, antral-predominant gastritis. With respect to duodenal ulcer disease, endoscopic visualization of the ulcer may be sufficient for diagnosis. Diagnostic confirmation of the presence of *H pylori* necessitates biopsy sampling of the gastric corpus and antrum. In contrast, the diagnostic evaluation of gastric ulcers requires biopsy specimens of the ulcer base and areas adjacent to gross ulceration to assess the histologic features for the presence of atrophic or neoplastic changes. Adjacent mucosa is evaluated directly for the presence of concomitant atrophy, dysplasia, intestinal metaplasia, or gastric adenocarcinoma.

Atrophic gastritis is characterized by glandular atrophy and likely represents a precursor state for gastric adenocarcinoma. Nonneoplastic mucosa adjacent to gastric carcinomas and lymphomas often displays atrophy, dysplasia, and intestinal metaplasia. *H pylori* organisms rarely are observed in association with regions of atrophic gastritis or gastric adenocarcinoma. A second form of gastric neoplasia, gastric MALT lymphoma, may be evaluated by histopathologic studies. Gastric marginal zone B-cell lymphomas (or MALT lymphomas) are highlighted histologically by the presence of lymphoepithelial lesions, prominent lymphoid follicles, and extensive zones of marginal B lymphocytes infiltrating the mucosa. *H pylori* organisms may be visualized adjacent to areas of low-grade gastric MALT lymphomas but are observed less commonly in association with diffuse large B-cell lymphomas (high-grade MALT lymphomas).

Mucosa adjacent to gastric ulcers or the gastric antrum likely harbor *H pylori*. Organisms rarely are visualized within gastric or duodenal ulcers. The presence of neutrophils in a mixed inflammatory pattern shown by H&E staining in patients with active gastritis is characteristic of *H pylori* gastritis. Organisms usually require visualization with special stains (eg, rapid Romanowsky, modified Giemsa, Genta, or silver stains). Fluorescent DNA probes recognizing ribosomal RNA (rRNA) sequences have been designed and used to detect *H pylori* in human gastric tissue by fluorescence in situ hybridization. Occasionally, sparse inflammation is observed despite the presence of low numbers of organisms visible with special stains, emphasizing the usefulness of special stains in the diagnosis of *H pylori* infection.

**Cell-Mediated Immune Response and Cytokines**

Multiple *H pylori* antigens stimulate the gastric epithelium to secrete multiple cytokines that initiate the inflammatory reaction within the gastric mucosa. Interleukin (IL)-8, leukotrienes, and complement activation products represent potent chemoattractants for neutrophils and lymphocytes. *H pylori* gene products activate host transcriptional regulatory factors such as NF–kappa B. The pathogenicity-associated island (PAI) of *H pylori* includes the *cagA* and *cagE* (*picB*) genes and is important for NF–kappa B activation in epithelial cells by a contact-dependent mechanism. NF–kappa B activation and subsequent gastric epithelial IL-8 induction occur in the presence of *H pylori* strains containing the 40-kilobase cag PAI. Type 1 *H pylori* strains containing the intact cag PAI represent especially potent inducers of mucosal IL-8. In particular, the PAI product of *cagE* seems to be an important inducer of IL–8 in patients with duodenal ulcer disease. Antigen-specific B and T lymphocytes accumulate within the lamina propria and the epithelium in response to *H pylori* infection. Chronic active gastritis is associated with an increased CD4/CD8 T-cell ratio within the gastric mucosa, due largely to the accumulation of CD4+ T-helper lymphocytes in the lamina propria. *H pylori* infection results in a Th1-predominant host immune response in the gastric mucosa, defined by the induction of interferon (IFN)-gamma and IFN-gamma–regulated
genes. A Th1-predominant immune response is generated with elevated levels of proinflammatory cytokines such as IL-12,\textsuperscript{49} IL-18,\textsuperscript{40} and tumor necrosis factor alpha.\textsuperscript{51,52} The severity of gastritis associated with \textit{H pylori} infection was correlated with mucosal expression of the tumor necrosis factor alpha subunit and IFN-gamma.\textsuperscript{52} The robustness of the mucosal Th1 response has been associated with progression to atrophic gastritis and gastric cancer,\textsuperscript{53} as supported by animal models. Immunoregulatory cytokines such as IL-10 also are induced in the gastric mucosa.\textsuperscript{49}

**Humoral Immune Response**

The presence of B lymphocytes and plasma cells in the gastric mucosa is evidence of an active humoral response in chronic infection. Sampling of gastric secretions from \textit{H pylori}–infected individuals reveals a robust mucosal antibody response, primarily of the IgA isotype.\textsuperscript{54} This response is consistent with the predominance of secretory IgA in the gastric secretions of healthy individuals. Secretory IgA anti–\textit{H pylori} antibodies also are found in saliva and breast milk.

Most patients infected with \textit{H pylori} produce a measurable systemic immune response, composed primarily of IgG.\textsuperscript{54,55} Serum IgA may be detected in fewer than half (range, 39%-82%) of infected patients, and serum IgM is found rarely. These findings are consistent with a chronic infection usually acquired in early childhood. Since few patients have been evaluated during the acute phase of infection, data about the nature of the acute antibody response and seroconversion from IgM to IgG are sparse. Seroconversion to IgG was demonstrated between 22 and 33 days after infection in one volunteer study.\textsuperscript{56} In naturally acquired infection,\textsuperscript{57} an initial serum IgM response was observed, and seroconversion of IgM to IgG was documented. Circulating anti–\textit{H pylori} IgG antibodies persist at constant levels for years during infection. Levels of IgG1, IgG2, and IgG4 subclasses typically are elevated, whereas IgG3 antibodies (associated with acute infections) are not detected.\textsuperscript{57} Autoimmune and corpus-predominant atrophic gastritis have many overlapping features. Antigastric antibodies are generated in a subset of infected individuals and include antiluminal and anticanalicular antibodies. A principal target of \textit{H pylori}–associated autoantibodies is the parietal cell proton pump, H\textsuperscript{+},K\textsuperscript{+}-ATPase.\textsuperscript{58}

**Serologic Evaluation of \textit{H pylori} Infection**

Serologic testing represents a primary screening approach for evaluation of \textit{H pylori} status in patients not immediately requiring EGD endoscopic studies. Pooled \textit{H pylori} antigens of high-molecular-weight surface-associated proteins, acid extracts, or whole cell lysates (sonicates) are used in most serologic assays. Various cytosolic and cell surface–associated proteins represent immunodominant antigens recognized by serum antibodies of infected individuals.\textsuperscript{59} Overall, the medians of the sensitivity and specificity for \textit{H pylori} serology kits have been reported as 92% and 83%, respectively.\textsuperscript{60} Serologic testing for serum IgG antibodies demonstrated performance superior to assays that measured total serum antibodies (serum IgA, IgG, and IgM) simultaneously or serum IgA alone.\textsuperscript{60} Performance varies significantly for commercial serologic kits, with top performers exceeding 90% in sensitivity and specificity and bottom performers having less than 90% in sensitivity and less than 80% in specificity.\textsuperscript{60} Additional studies report variable sensitivities (86%-100%) and specificities (76%-98%) of assays measuring anti–\textit{H pylori} serum IgG.\textsuperscript{61,62} It is important to note that positive (95%-100%) and negative (84%-89%) predictive values for serology were comparable to those of histologic examination, rapid urease testing, and urea breath testing (UBT).\textsuperscript{63} Enzyme-linked immunosorbent assay serologic testing had the lowest cost per correct diagnosis, but overall accuracy was lower than stool antigen testing or UBT.\textsuperscript{64} Patients infected with “Helicobacter heilmannii” usually had negative results in anti–\textit{H pylori} IgG assays.\textsuperscript{65} Serum IgG immunoassays have markedly reduced sensitivities (50%-60% vs >90%) in HIV–1–infected patients.\textsuperscript{66} Therefore, a negative result in this setting should be confirmed by UBT or gastric biopsy.

To facilitate in-office point-of-care testing for \textit{H pylori} infection, rapid serum and whole blood IgG immunoassays have been developed by several manufacturers (eg, QuickVue \textit{H pylori} Test, Quidel, San Diego, CA; FlexSure HP, SmithKline Diagnostics, Palo Alto, CA). A rapid latex agglutination test format (Pyloriset, Orion Diagnostics, Espoo, Finland) also is available to detect serum anti–\textit{H pylori} IgG. Such immunoassays produce a qualitative result in 4 to 10 minutes using heparinized whole blood or capillary blood specimens. Compared with serum enzyme immunoassays, whole blood serologic assays have reduced sensitivities (80%-90% vs >90%) with comparable specificities (70%-80%) and lower overall accuracy.\textsuperscript{67,68} Whole blood card-based immunoassays represent acceptable approaches for diagnostic screening in the outpatient setting, assuming that negative results are confirmed by laboratory enzyme-linked immunosorbent assay–based serum IgG testing, fecal antigen testing, or UBT.

Assays to detect anti–\textit{H pylori} IgA in serum samples have reduced and variable sensitivities (39%-82%) compared with serum IgG immunoassays. Serum IgA studies may be useful in testing of symptomatic individuals with equivocal or negative IgG findings. In one study, more than 7% of patients with negative serum IgG results were found to have detectable anti–\textit{H pylori} serum IgA and symptoms consistent...
with *H pylori* infection. Of 6 patients with IgA-positive, IgG-negative results on the IgA immunoassay from HYCOR Biomedical (Irvine, CA), 5 had peptic ulcers documented by endoscopy. Salivary and urinary antibody assays have shown limited usefulness.

### Urease and *H pylori*

*H pylori* produces copious amounts of cell surface–associated urease as a catalytically active heterodimer. It is estimated that urease comprises more than 5% of bacterial protein and at least 1 subunit (UreB) comprises an immunodominant antigen. Assumee of urease activity is the basis for detection of the presence of the organism by biopsy-based rapid urease testing (eg, CLOtest, Ballard Medical/Kimberly-Clark, Draper, UT; hpfast, GI Supply, Camp Hill, PA) or UBT. *H pylori* urease is secreted, becomes cell surface–associated, and elevates the local intragastric pH by catalyzing urea breakdown and ammonia production. Urease also has an important role in the bacterial nitrogen cycle. Urease-negative mutants fail to cause persistent infection in animal models of *Helicobacter* infection, indicating that urease represents an important factor for survival of *H pylori* in the stomach.

### Rapid Urease Testing

The first application of *H pylori* urease activity for diagnosis was rapid urease testing of gastric biopsy material. Urease catalyzes the hydrolysis of urea into ammonia and carbamate. The net effect of ammonia production is increased local pH. Biopsy samples are placed in an agar gel or paper strip containing a pH indicator. If organisms are present in sufficient numbers in the antral biopsy sample, a color change will occur as a result of urea breakdown and ammonia production by *H pylori* urease. Commercial rapid urease tests include the agar gel–based tests (eg, CLOtest; hpfast) or paper strip tests (eg, PyloriTek, Horizons International, Ponce, Puerto Rico). Sensitivity of detection depends on organism load in the mucosal biopsy specimen and the number of biopsy samples. Optimally, biopsy samples of the corpus and antrum should be obtained for rapid urease testing. One antral biopsy specimen may be sufficient for rapid urease testing in patients with duodenal ulcer disease, but the location of biopsy sampling is important and should be based on endoscopic evaluation. The sensitivity of rapid urease testing exceeds 95% in patients with gastric ulcer disease if the corpus is sampled, whereas the sensitivity is diminished markedly if only antral samples are obtained from these patients. Rapid urease tests enable convenient detection of *H pylori* infection within 3 hours in most cases, although the agar gel–based tests (eg, CLOtest, hpfast) usually require 24 hours of incubation for maximal sensitivity and specificity.

Biopsy-based rapid urease testing demonstrates excellent specificity (range, 93%-100%) but less than impressive sensitivity (range, 89%-98%). Sensitivity of rapid urease testing with single biopsy specimens typically ranges between 85% and 90%. Because the sensitivity and specificity of rapid urease tests approach or exceed 90%, rapid urease tests may be cost-effective screening methods to avoid unnecessary histopathologic evaluation. False-positive results of follow-up rapid urease tests may reflect effective treatment with insufficient posttherapy periods, so that adequate follow-up periods are recommended (at least 6 weeks). Rapid urease testing is less sensitive in the pediatric setting, and special stains or noninvasive direct detection may be important to rule out the diagnosis. Finally, urease-negative *Helicobacter* organisms have been reported in cases of gastritis with negative results of rapid urease testing, so the possibility of infection with organisms other than *H pylori* should be considered in patients with refractory disease and negative serologic and urease testing results.

### Urea Breath Testing

Noninvasive UBT has excellent sensitivity and specificity for the diagnosis and assessment of patients after antimicrobial therapy (both >95%) compared with invasive methods such as culture, histopathologic examination, or rapid urease testing. The US Food and Drug Administration has approved both carbon 13 (13C)– and carbon 14 (14C)–based urea breath tests for the diagnosis of *H pylori*. However, the higher cost per assay makes it difficult to recommend UBT instead of serology for routine screening of *H pylori* infection in patients with uncomplicated disease. Invasive tests such as histopathologic examination or the rapid urease tests require EGD endoscopy, with total costs in the United States usually exceeding $1,000. Because of its noninvasive nature and usefulness soon after therapy, UBT is recommended for follow-up of patients who continue to be symptomatic despite antimicrobial therapy. UBT may confirm eradication of *H pylori* within 6 weeks after therapy compared with a 6 to 12 month follow-up period with serologic testing. Asymptomatic patients who have experienced upper gastrointestinal bleeding may warrant posttherapy monitoring by UBT. If effective therapy of peptic ulcer disease is delayed and *H pylori* is not eradicated, patients remain at risk for serious complications such as hemorrhage and perforation.

The urea breath test is performed by having the patient consume a solution of isotopically labeled urea dissolved in aqueous solution followed by breath collection 30 to 60 minutes later. An enhanced version that includes a citric acid test meal shortened time to breath collection (15 minutes)
with $^{13}$C-urea. Breath samples are transferred to evacuated sample tubes for subsequent measurement of exhaled, isotopically labeled carbon dioxide. Both the $^{14}$C and $^{13}$C isotopes have been used for clinical testing, although the stable isotope, $^{13}$C, is more widely used in the United States. The use of this method for the diagnosis of $H$ pylori infection was first documented in 1987, and its clinical usefulness has been demonstrated in multiple studies.

Antigen Detection

Direct fecal antigen detection of $H$ pylori has been approved by the US Food and Drug Administration for diagnosis and follow-up testing. Meridian Bioscience (Cincinnati, OH) developed a commercial kit (Premier Platinum HpSA) for the rapid, noninvasive detection of $H$ pylori antigens by enzyme immunoassay. $H$ pylori antigens from fresh human fecal specimens are detected by polyclonal antibodies adsorbed to microwells. The sensitivity and specificity of fecal antigen detection were approximately 89% and 94% to 95%, respectively, in multiple studies. Fecal antigen detection and UBT are recommended noninvasive approaches for confirmation of $H$ pylori infection in pediatric patients because serologic tests are less reliable (especially in children younger than 5 years) and are useful only for screening in this population.

DNA Detection: Polymerase Chain Reaction–Based Approaches

Strategies for polymerase chain reaction (PCR)-based detection of $H$ pylori have included multiple genetic targets with varying levels of sensitivity and diagnostic accuracy. Comparisons of different target sequences in gastric biopsy specimens yielded different positive and negative predictive values. The $\text{glmM (ureC)}$ gene encoding phosphoglucomutase yielded superior overall results for detection of $H$ pylori by PCR. The $\text{vacA}$ gene encoding the putative virulence factor, vacuolating cytotoxin, yielded excellent results with sensitivity of PCR detection exceeding 99%. Maeda et al reported a PCR assay based on 23S ribosomal DNA (rDNA) target sequences that yielded specific amplification of $H$ pylori from gastric biopsy specimens and that may be useful when combined with molecular resistance testing. Other investigators have amplified $H$ pylori DNA from human saliva, gastric juice, and feces, although with diminished sensitivities. Real-time detection and quantitation of $H$ pylori DNA in gastric biopsy specimens was performed with the $\text{glmM}$ gene as the target and represents an important development in molecular diagnostics for $H$ pylori infection.

Microbiologic Culture and Susceptibility Testing

$H$ pylori is a fastidious organism that requires an enriched transport medium if the biopsy specimen is not plated for culture within 2 hours. Because optimal transport media are not available commercially and microbiologic plating usually is not performed in the endoscopy unit, routine cultures have not acquired general acceptance in the United States. The increasing prevalence of drug-resistant $H$ pylori infections may necessitate susceptibility testing. Infection with $H$ pylori strains resistant to clarithromycin or metronidazole is associated with an increased incidence of treatment failure.

Emergence of antimicrobial-resistant $H$ pylori represents an important challenge for the treatment of currently infected patients. Studies indicate worrisome levels of resistance to the most important antimicrobial agents, metronidazole and clarithromycin. In Wisconsin, the prevalence of macrolide-resistant $H$ pylori among infected individuals increased from 4% during the 1993-1994 period to 12.6% during the 1995-1996 period. In multiple centers throughout the United States, levels of metronidazole resistance approached 40%, and the prevalence of clarithromycin resistance exceeded 10%. Resistance to either metronidazole or clarithromycin has been associated with markedly reduced eradication rates after extended treatment with multiagent regimens. Patients who do not respond to antimicrobial therapy and have documented persistent infection may require antimicrobial susceptibility testing.

Antimicrobial susceptibility testing methods such as microbroth dilution, disk diffusion, E-test, and agar dilution have been used to assess antimicrobial resistance in $H$ pylori. The National Committee for Clinical Laboratory Standards (NCCLS) recommends an agar dilution standard for $H$ pylori susceptibility testing. Mueller-Hinton agar base with 5% aged sheep blood and incubation for 72 hours at 35°C were selected by the NCCLS for susceptibility testing by agar dilution. Resistance in vitro to clarithromycin by $H$ pylori is clinically relevant, and a minimum inhibitory concentration breakpoint of 1 µg/mL is recommended by NCCLS.

Molecular diagnostics may enable the rapid detection of antibiotic resistance determinants in $H$ pylori. More than 90% of clarithromycin-resistant isolates contain either of 2 point mutations in the 23S rRNA gene. PCR-based detection of mutations associated with macrolide resistance in $H$ pylori may be practical and clinically useful. Point mutations in the 23S rRNA gene confer stable macrolide resistance, and either of 2 predominant mutations may be detected by PCR amplification and restriction digestion or oligonucleotide ligation. Gastric biopsy specimens may
be rescued from CLO tests even if samples have been kept at room temperature for up to 8 weeks after testing. This approach may be useful for molecular diagnosis in patients with disease that is refractory to therapy who are not candidates for follow-up endoscopy. Rescued biopsy specimens may be used for detection of resistance mutations by PCR–restriction fragment length polymorphism approaches. PCR amplification and reverse hybridization is a successful strategy to detect multiple 23S rDNA mutations in mixed populations and yields excellent sensitivity of detection by colorimetric approaches. Real-time PCR approaches have been described for fluorescence detection of 23S rDNA mutations conferring macrolide resistance.

**Antimicrobial Therapy for H pylori Infection**

Multiple therapeutic regimens have been demonstrated to effectively cure patients of *H pylori* infection. Metronidazole or clarithromycin must be included to obtain eradication rates exceeding 90%. The highly effective MOC regimen includes metronidazole, omeprazole, and clarithromycin and may be administered for 7 to 14 days, yielding greater than 90% eradication. No single agent has been shown to be effective for curing infection in the majority of patients. Because of the increasing problem of antimicrobial resistance, extended (10-14 day) proton pump inhibitor–based regimens with at least 2 drugs (preferably 3 or 4) should be used for treatment. Fourteen-day bismuth-based regimens with either tetracycline or amoxicillin and metronidazole represent cost-effective options in developing countries and effective regimens in areas with documented low levels of resistance. Proton pump inhibitors (eg, omeprazole) or H₂-antagonists are included routinely in multiagent regimens to accelerate ulcer healing and ameliorate symptoms.

**Summary of Diagnostic Strategies**

Several diagnostic strategies are available. Routine screening of asymptomatic patients is not recommended. Initially, patients with uncomplicated dyspeptic disease (especially patients younger than 50 years) should undergo serologic testing for the presence of anti-*H pylori* antibodies in serum or whole blood. If the serum IgG result is negative and the clinical index of suspicion remains high, a urea breath test or fecal antigen test for direct detection and diagnostic confirmation would be recommended. Serum IgA testing also may be considered. If “alarm features” such as older age, evidence of gastrointestinal bleeding, or weight loss are present, EGD endoscopy should be performed, and obtaining a gastric mucosal biopsy specimen must be considered. Biopsy specimens may be submitted for rapid urease testing or histologic examination. Antral and corpus biopsy specimens are recommended for maximal sensitivity of detection, and tissue submitted for histopathologic examination should undergo special stains to enable organism visualization. Patients with evidence of ulcer disease and *H pylori* infection should be treated promptly with antimicrobial therapy to eradicate *H pylori*. Since long-term consequences of *H pylori* infection include substantial risks for peptic ulcer disease or gastric neoplasia, patients diagnosed with *H pylori* infection always should be considered for treatment and follow-up testing depending on the clinical manifestations.

Patients whose symptoms persist after therapy should be evaluated at 6 to 8 weeks after therapy by UBT or fecal antigen detection. Elderly patients with documented ulcer disease or previous complications should be monitored routinely by follow-up UBT, fecal antigen detection, or EGD endoscopy with biopsy. Patients with disease that is refractory to therapy may undergo repeated endoscopy for antimicrobial susceptibility testing. Alternatively, if follow-up endoscopy is unnecessary, biopsy specimens used in rapid urease tests may be rescued for molecular resistance testing. Molecular data may be used to formulate rational modifications in anti–*H pylori* treatment regimens.

**References**


