RESEARCH ARTICLE

Evaluation of Invasive and Noninvasive Methods for the Diagnosis of Helicobacter Pylori Infection

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Abstract

Objective: The present study was conducted to evaluate invasive and noninvasive diagnostic methods for detection of Helicobacter pylori (H. pylori) in patients admitted with dyspeptic complaints and to compare sensitivities and specificities.

Method: Sets of four gastric biopsy specimens were obtained from a total of 126 patients included in the study. The presence of H. pylori was determined by invasive tests including culture, rapid urease test, polymerase chain reaction (PCR) and histopathology. Among noninvasive tests, urea breath test, serological tests and enzyme-linked immunosorbent assay (ELISA) were performed.

Results: H. pylori was isolated in 79 (62.7%) gastric biopsy cultures, whereas positivity was concluded for 105 (83.3%) patients by rapid urease test, for 106 (84.1%) by PCR, for 110 (87.3%) by histopathology, for 119 (94.4%) by urea breath test, and for 107 (84.9%) by ELISA. In the present study, the culture findings and histopathological examination findings were accepted as gold standard. According to the gold standard, urea breath test had the highest sensitivity (96.5%) and the lowest specificity (30%), whereas culture and histopathology had the highest specificities (100%).

Conclusion: The use of PCR invasively with gastric biopsy samples yielded parallel results with the gold standard. PCR can be recommended for routine use in the diagnosis of H. pylori.

Keywords: Helicobacter pylori, diagnostic tests, polymerase chain reaction

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Introduction

H. pylori, which colonizes the gastrointestinal tract of nearly half of the world’s population, is a spiral-shaped, gram-negative microorganism causing local inflammation in the stomach and duodenum and inducing humoral immune response systemically (Ozden, 2006; Sanders et al., 2006). Recently, H. pylori is considered a causative agent in the development of non-ulcer dyspepsia, peptic and gastric ulcers, chronic gastritis, gastric carcinoma, and mucosa associated lymphoid tissue (MALT)-type lymphoma. H. pylori was classified as a class I human carcinogen by World Health Organization in 1994 (Asaka, 2002; WHO, 1997). In February 1994, National Institutes of Health proposed that patients infected with H. pylori should be absolutely treated with antibiotics (Thamer et al., 1998).

Numerous invasive and noninvasive methods have been developed for the diagnosis of H. pylori and many of them have been implemented in practice. Invasive methods include direct microscopic examination, culture, rapid urease test, histopathological examination, and molecular evaluation of biopsy samples; noninvasive methods include urea breath test, serological tests, fecal antigen test, measurement of 13C in blood and stool samples, molecular evaluation of samples other than biopsy samples.

There is no single test serving as a gold standard for the diagnosis; however, culture and histopathological analysis of biopsy samples have been accepted as the gold standard methods. A novel noninvasive, cheap, easily applicable, rapid method with high sensitivity and specificity has not been established yet (Dunn et al., 1997; Li et al., 1996; Laheij et al., 2000). Invasive and noninvasive methods have both advantages and disadvantages. The choice of the diagnostic method depends on complaints and age of the patients, as well as physical examination findings, previous history of endoscopy, family history of gastric cancer, whether the patient had received treatment or not, H. pylori seroprevalence in the population, and preference of the patients and physicians. Generally accepted approach is the use of two or three biopsy-based tests as a gold standard (Laheij et al., 2000; Ogata et al., 2001; Vinette et al., 2004).

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Materials and Methods

In the present study, invasive methods such as rapid urease test, culture, histopathology, and polymerase chain reaction (PCR) and noninvasive methods such as urea breath test and serological tests were used to diagnose *H. pylori* in endoscopic biopsy samples obtained from patients with dyspeptic complaints; and it was aimed to compare the sensitivity and specificity of these invasive and noninvasive methods in the diagnosis.

**Materials and Methods**

This study was carried out in 4 months at the Ankara Training and Research Hospital Microbiology Laboratory. One-hundred twenty-six patients, who were admitted to Outpatient Clinic of Internal Diseases with dyspeptic complaints and in whom endoscopy was indicated, were included in the study. The study group comprised patients not using antibiotics in the last one month, those not using proton-pump inhibitors (PPIs) for the last 14 days, and those not using H2 receptor antagonist or antacids for the last 24 hours.

**Samples**

Totally four biopsy specimens (two antral and two corpus biopsy specimens) were obtained from the patients in whom esophagogastroduodenoscopy was performed following at least eight hours of fasting. The first biopsy sample was placed in a glass container containing 1 mL sterile saline (0.9% NaCl); the second biopsy sample was placed in a glass container containing 1 mL 2% Christensen’s urea medium for culture; the third biopsy sample was placed in a 2 mL eppendorf tube containing 1 mL nuclease-free distilled water for PCR. Following 8 hours of fasting, 7 mL blood samples of the patients were drawn into serum separator tubes with gel for serological tests. Breath samples were collected from patients for urea breath test.

**Invasive Methods**

**Rapid urease test**

The samples, which were placed in tubes containing rapid urease medium and immediately transferred to the laboratory, were examined at endoscopy unit. The color of the medium changed from yellow to dark pink and fuchsia color was accepted as a positive result. If no change was observed in the color of the medium, samples were incubated at 35°C for 24 hours. The samples were re-evaluated at the 2nd and 24th hours in terms of positivity.

**Culture**

Brain Heart Infusion (BHI) agar (Oxoid, England) containing 10 mg/L vancomycin, 5 mg/L cefsulodin, 5 mg/L amphotericin-B, 5 mg/L trimethoprim (*H. pylori* selective medium, Oxoid, England) and 10% defibrinated horse blood was used. The biopsy samples, which were transferred to laboratory in glass tubes containing 1 mL sterile saline (0.9% NaCl), were cut into pieces using scalpel in sterile glass petri dishes. The samples were homogenized and then inoculated to BHI agar. The plates were placed in a 3.5 L jar with CampyGen (Oxoid, England) and the jar was closed within one minute. The plates were incubated for 7 days at 35°C. At the end of the incubation period, the grey, translucent colonies 1-2 mm in diameter with weak beta-hemolysis were considered suspicious for *H. pylori* and examined by conventional methods. Pure growth was obtained, since selective medium was used. The bacteria were identified by Gram staining and by performing urease, oxidase and catalase tests. Curved, S-shaped or seagull-wing-shaped appearance in Gram staining and positive urease, oxidase and catalase tests were defined as *H. pylori* positivity.

**Polymerase Chain Reaction for the detection of *H. Pylori* in biopsy samples**

In-house nested PCR assay was used to detect *H. pylori* DNA. DNA extraction was performed by the phenol-chloroform method. The primers were 5'-AACGGTCCTAAGGGATCGAA-3' and 5'-ACACTCAACTTGCGATTCC-3' for the first-step amplification 408 base pairs (bp) product of *H. pylori* 23S rRNA, and the primers were 5'-ATGAATGGCGTACAGGAT-3' and 5'-TGCTAAGTTGTAGTAAAGGT-3' for the second-step amplification 126 bp product of *H. pylori* 23S rRNA.

PCR reaction mixture was prepared in 5 μL eppendorf tubes by mixing 5 μL 10X PCR buffer, 0.5 μL dNTPs (10 mM), 0.2 μL Taq polymerase (5 U/μL), 2 μL magnesium (25 mM), primers 0.5μL for each (100 pmol/μL), and 36.3 μL nuclease-free water. Finally, 5 μL DNA samples were added to the PCR reaction mixture, and the tubes were placed in a thermal cycler device (Techne, England) for first-step amplification. *H. pylori* NCTC 11637 reference strain was used as positive control and nuclease-free water was used as negative control.

The amplification conditions for the first-step amplification product were: one cycle of initial denaturation at 94°C for 5 min, 30 cycles of denaturation at 94°C for 30 sec, annealing at 55°C for 45 sec, one cycle of elongation at 72°C for 60 sec, and one cycle of final elongation at 72°C for 5 min. A 5 μL DNA sample from the obtained amplification product was added to the PCR mixture (45 μL) in 0.5 mL eppendorf tubes and the tubes were placed in thermal cycler device (Techne, England). The amplification conditions for the second-step amplification product were: one cycle of initial denaturation at 94°C for 3 min, 30 cycles of denaturation at 94°C for 20 sec, annealing at 55°C for 35 sec, elongation at 72°C for 45 sec, and one cycle of final elongation at 72°C for 5 min.

Amplified PCR products, positive and negative control samples and DNA marker (Gene Ruler 100bp DNA ladder, MBI Fermentas, Lithuania) were transferred to wells and separated by agarose gel electrophoresis; direct current was applied at a voltage of 150 V for 35-40 min. Results were evaluated in terms of the presence of first-step amplification 408 bp product and second-step amplification 126 bp product with a UV transilluminator (Vilber Lourmat, France).

**Histopathological Examination**

6-μm in-thick sections were obtained from the specimens that were transferred to the pathology laboratory in 10% neutral formalin solution. The sections
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There was no significant difference between female and male patients in terms of *H. pylori* positivity (p>0.05).

Patients with *H. pylori*-positive status is shown in Figure 2 according to the age group (Figure 2).

The most frequent endoscopic diagnosis was antral gastritis (Table 1).

**Results**

In the present study, the culture findings and histopathological examination findings were accepted as gold standard. The detection of *H. pylori* at least one of the two tests was accepted as *H. pylori* positivity. Negative results in both culture and histopathology were accepted as *H. pylori* negativity.

The age range of the 126 patients (85 females (64.4%) and 41 males (32.6%)) was 14 to 74 years. The mean ages of female and male patients were 37.1±13.1 and 38.8±13.1 years, respectively (Figure 1).

No statistically significant difference was determined between male and female patients in terms of age (p>0.05).

**Noninvasive Methods**

**Serological Evaluation**

Serum antibody levels were measured quantitatively with a micro enzyme-linked immunosorbent assay (ELISA) device (Alisei, Italy) using a commercial ELISA kit (Radim, Italy) to detect anti-*H. pylori* IgG and anti CagA IgG antibodies.

**Urea Breath Test**

By paying attention that patients did not use antibiotics and bismuth within the last one month, PPIs within the last 14 days, and antacids and H2 receptors within the last 24 hours, a capsule (HeliCap) containing 1 μCi 14C-labeled urea and citric acid was administered perorally with 50 mL water to the patients following at least six hours of fasting. Ten minutes after administration of the capsule, the patients exhaled into breath cartridge (Heliprobe Breathcard, Sweden) for five minutes until the color changed from orange to yellow. The cartridge was placed in an analyzer (Heliprobe-analyzer, Sweden) and activity count was performed for 250 sec. The labeled CO2 collected in the cartridge was expressed as counts per min (cpm). The result was calculated automatically by the device and graded as grades 0, 1, and 2.

**Culture**

Of 126 patients, *H. pylori* was isolated from gastric biopsy cultures of 79 (62.7%) patients; however, *H. pylori* could not be isolated in 47 (37.3%) patients. *H. pylori* was detected by histopathology in 37 of 47 patients in whom culture results for *H. pylori* was negative; these results were interpreted as false negative results for culture. A significant difference was found between the culture method and the gold standard in terms of the rate of *H. pylori* positivity (p=0.0001).

**Histopathological Examination**

While *H. pylori* positivity was determined in 110

Table 1. Distribution of the Study Patients According to Endoscopic Diagnosis

| Endoscopic diagnosis      | n (%) |
|--------------------------|-------|
| Panagastrosis             | 11 (8.7) |
| Antral gastritis          | 60 (47.6) |
| Antral gastritis + duodenitis | 16 (12.7) |
| Antral gastritis + esophagitis | 17 (13.5) |
| Esophagitis               | 2 (1.6) |
| Peptic ulcer              | 17 (13.5) |
| Normal                    | 3 (2.4) |
| Total                     | 126 (100) |
Table 2. The Sensitivity, Specificity, Positive and Negative Predictive Values of the Tests Performed in the Study According to Gold Standard

| Tests                | Sensitivity (%) | Specificity (%) | PPV (%) | NPV (%) |
|----------------------|-----------------|-----------------|---------|---------|
| Culture              | 68.1            | 100             | 100     | 21.2    |
| Rapid urease test    | 87.9            | 70              | 97.1    | 33.3    |
| PCR                  | 88.7            | 70              | 97.1    | 35      |
| Histopathology       | 94.8            | 100             | 100     | 62.5    |
| Urea breath test     | 96.5            | 30              | 94.1    | 42.8    |
| ELISA                | 86.2            | 40              | 94.3    | 20      |

(87.3%) patients by histopathological examination of biopsy samples, the presence of *H. pylori* was not determined in 16 (16.6%) patients. *H. pylori* positivity was determined by culture in 6 of 16 patients in whom *H. pylori* negativity was determined by histopathology; these results were interpreted as false negative results for histopathological examination. No significant difference was found between the histopathological examination and the gold standard in terms of the rate of *H. pylori* positivity (p=0.3).

**Rapid Urease Test**

*H. pylori* positivity was determined in biopsy samples of 105 (83.3%) patients and *H. pylori* negativity was determined in biopsy samples of 21 (16.7%) patients by rapid urease test. No significant difference was found between the rapid urease test and the gold standard in terms of the rate of *H. pylori* positivity (p=0.054).

**Urea Breath Test**

Of 126 patients, *H. pylori* positivity was determined in 119 (94.4%) patients, whereas *H. pylori* negativity was determined in 7 (5.6%) patients by urea breath test. No significant difference was found between the urea breath test and the gold standard in terms of the rate of *H. pylori* positivity (p=0.62).

**Polymerase Chain Reaction (PCR)**

*H. pylori* positivity was determined in 106 (84.1%) patients and *H. pylori* negativity was determined in 20 (15.9%) patients by PCR of biopsy samples. No significant difference was found between the PCR method and the gold standard in terms of the rate of *H. pylori* positivity (p=0.07).

**Enzyme-Linked Immunosorbent Assay (ELISA)**

*H. pylori* positivity was determined in 107 (84.9%) patients and *H. pylori* negativity was determined in 19 (15.1%) patients by ELISA in serum samples of 126 patients. No significant difference was found between the ELISA and the gold standard in terms of the rate of *H. pylori* positivity (p=0.11). In addition, anti-*H. pylori* CagA IgG antibodies were measured in the patients’ sera using ELISA method. Of the 107 patients in whom anti-*H. pylori* IgG was positive, 75 (70%) patients were positive for anti-*H. pylori* CagA IgG (Table 2).

**Discussion**

The excitement beginning with isolation of *H. pylori* from gastric biopsies by Warren and Marshall in 1982 has increasingly continued after the important role of this bacterium in the etiology of gastric cancer has been established; thus, the interest in *H. pylori* has increased (Ozden, 2006). The association of gastric cancer, one of the most frequent causes of death worldwide, with a treatable etiological factor have led to a profound impact on the researchers in the field of gastro-oncology and brought the 2005 Nobel Prize to the researchers who isolated the bacteria and proved its association with gastric diseases (Ozden, 2006; Marshall and Warren, 1984). *H. pylori* infection is acquired in early childhood, and colonization occurs in the stomach. The earlier the infection starts, the risk of complications associated with *H. pylori* increases (Asaka, 2002; Dunn et al., 1997; Konturek, 2003; Brown, 2001; Yang et al., 2002).

Accurate and suitable diagnosis and treatment of this infection affecting human health to such a serious extent, deteriorating quality of life of all age groups, and even causing death is of importance. For this purpose, many diagnostic methods have been developed and some of them have been implemented in practice. Each method has both advantages and disadvantages due to different reasons such as discomfort to patients, side effects, costs, duration of the tests, workforce, and requirements of special conditions.

In the recent consensus reports, the use of noninvasive methods, particularly urea breath test and fecal antigen tests, have been suggested for the detection and treatment of *H. pylori* in patients under the age of 45 years with dyspeptic complaints and without alarm symptoms in the populations with high seroprevalence rates, like in Turkey. The use of invasive methods requiring endoscopic biopsy has been accepted in the presence of alarm symptoms (namely epigastric pain, severe vomiting, unexplained weight loss, unexplained anemia), in subjects over the age of 45 years, in case of impossibility to eradicate the bacteria despite treatment, and in the presence of family history of gastric cancer (Ozden, 2006; Mitchell and Megraud, 2002; Ellidokuz et al., 2003). Histopathology among invasive methods and urea breath test and ELISA among noninvasive methods are the most frequently used methods in the world and in Turkey (Ozden, 2006; Dunn et al., 1997; Malaty et al., 2002; Ozden et al., 2004; Weiss et al., 1994).

In the studies evaluating diagnostic methods for *H. pylori*, the use of biopsy-based invasive methods has been accepted as gold standard. Gold standard is determined by the combined use invasive methods, particularly culture and histopathology, in some studies, whereas
Since resistance is encountered in one of every four patients diagnosed with noninvasive methods and treated in the populations in which the rate of clarithromycin resistance is over 25%, like in Turkish population, sensitivity tests become important. When endoscopy is performed on the patients who are treated after the diagnostic test but in whom symptoms do not regress, obtaining biopsy specimens for sensitivity and culture tests in addition to histopathological examination appears to be the best suitable approach (Asaka, 2002; Thamer et al., 1997; Dunn et al., 1997; Brown, 2000; Baglan et al., 2005; Baglan et al., 2006). Since histopathological examination is based on direct observation of bacteria in the samples by means of stains, it is a method with a high sensitivity and specificity rates (Wong et al., 2014; Dunn et al., 1997; Weiss et al., 1994). In our study, histopathological examination was evaluated as a gold standard together with culture. It was found to be the test with the second highest sensitivity after urea breath test. The sensitivity and specificity of histopathological examination were calculated to be 94.8% and 100%, respectively. In the study performed by Vinette et al. on pediatric patients, the sensitivity and specificity of histopathological examination was reported to be 100% and 97.8%, respectively (Vinette et al., 2004). In their study Kobayashi et al. reported the sensitivity to be 99% and specificity to be 94% for histopathological evaluation (Kobayashi, 2002). False negative results for the histopathological examination were considered to depend on patchy localization pattern of the bacteria in the gastric epithelium.

As rapid urease test is based on biopsy, it has been accepted as a gold standard test together with culture or histopathology in many studies (Laheij et al., 2000; Vinette et al., 2004; Kaya et al., 2007). In addition to being cost-effective, rapid urease test has some advantages such as not requiring special conditions and providing results in a short period of time (within one hour) (Dunn et al., 1997).

In the present study, H. pylori positivity was determined in 105 patients by rapid urease test and H. pylori negativity was determined in 21 patients. The sensitivity, specificity and PPV of the rapid urease test were found to be 87.9%, 70% and 97.1%, respectively. No significant difference was determined between the rapid urease test and gold standard in terms of the rates of H. pylori positivity (p=0.054). Two of the three false-positive results were also determined to be positive by urea breath test. Since both tests are based on urease enzyme, it was considered that this result could be due to the contamination by other bacteria producing urease. In their study, Brandi et al., (2006) performed urea breath test, rapid urease test, culture and histopathological examination on biopsy samples and determined that urease-positive bacteria other than H. pylori led to false positive results for rapid urease and urea breath tests.

Polymerase chain reaction technique can be used for the determination of diagnosis and antibiotic sensitivity to detect bacteria and resistant genes. In addition, it can be used to detect genetic differences among H. pylori species in epidemiological studies. In the present study, nested PCR method used for detecting a target sequence located in the 23S rRNA showed a good correlation.
with gold standard methods. Of the 126 samples, *H. pylori* positivity was determined in 106 and *H. pylori* negativity was determined in 20. Three false positive and 13 false negative results were obtained. The fact that PCR is a method prone to contamination, and the possibility of determining *H. pylori* positivity due to bacteria colonization in oral region and dental plaques and amplification of DNA from dead bacteria affect the reliability of PCR technique.

Three false positive results determined in our study were lower than the rates of false positive results found in the studies performed using PCR technique. The target gene region and the PCR technique used affect the reliability of the test in the studies utilizing PCR technique. In the PCR studies, in which a target sequence located in the 16S rRNA has been selected, the rates of false positive results have been found to be high (Trebesius et al., 2000; Bulut et al., 2006). In their study, Lu et al. compared five separate primer pairs and accepted culture as the gold standard method. They reported the sensitivity of 16S rRNA to be 100%; however, they stated that it was not reliable, since the NPV of the technique was determined to be 46% (Lu et al., 1999). Furthermore, the PPV and NPV of the PCR of ureC (glmM) gene was determined to be 100% and 96% and this test was reported as the PCR test with the highest specificity and sensitivity (Lu et al., 1999).

Among the biopsy-based tests in the present study, the best result was obtained by PCR, following histopathology. No significant difference was determined between the gold standard tests and PCR in terms of the rate of *H. pylori* positivity (p=0.07). In addition to the fact that 23S rRNA was targeted, performing nested PCR may have increased the sensitivity and specificity. In the studies determining *H. pylori* DNA using PCR, false negative results may also be obtained (Weiss et al., 1994). In our study, 13 false negative results were determined by PCR. False negative results detected by PCR in the present study was considered to be resulted from the patchy localization of the bacteria in stomach and interaction of sample with inhibitors as the PCR method used was conventional.

Serological methods are mostly used in population-based seroprevalence studies, as these tests do not allow the diagnosis of active infections (Göral et al., 2006; Akin et al., 2004). In the populations with high seroprevalence rates, however, serological methods have been suggested to be used as the first method if urea breath test and fecal antigen test are not available (Ozden, 2006). Regarding the high virulence of strains, the presence of anti-CagA antibodies is correlated with CagPAI gene detected by molecular studies.

In the present study, the sensitivity, specificity, PPV and NPV of the ELISA method used to detect anti-*H. pylori* IgG was determined to be 86.2%, 40%, 94.3% and 20%, respectively. As specificity and NPV of this method were determined as low as 40% and 20%, respectively, we considered that it is not a test to be preferred for diagnostic purposes; it can be used as a noninvasive test when urea breath test is not available. In the study by Li et al. the sensitivity and specificity of ELISA method used for the determination of serum IgG positivity was determined to be 97% and 94%; these values were higher than those determined in our study (Li et al., 1996). In the study by Vinette et al. (2004) in which *H. pylori* was investigated in gastric biopsy samples of 101 pediatric patients, the sensitivity and specificity of serology was found to be 69.2% and 78.2%, respectively. They indicated that this may have been depended on inadequate antibody response at pediatric age group and that the use of serological methods for diagnosis of *H. pylori* is not suitable in pediatric patients (Vinette et al., 2004).

In our study, anti-CagA IgG positivity was determined in 75 patients of 107 patients in whom anti-*H. pylori* IgG positivity was determined; a positivity rate of 70% was calculated for all patients. This rate found in the present study was in parallel to the rates reported in other studies conducted in Turkey (Ozden et al., 2004; Sarıbasak et al., 2004). Ryan et al., (2001) investigated clarithromycin resistance and virulence genes in gastric biopsy samples of 50 patients by PCR and determined that all *H. pylori* positive strains were VacA-positive and 76% of *H. pylori* positive strains were CagA-positive (1).

Urea breath test is accepted as the most suitable method to diagnose *H. pylori* and for the control of eradication in adult patients with dyspeptic complaints, since urea breath test is easy to use, provides results in short period of time (like 20-30 minutes), and has a quite high sensitivity (Ozden, 2006; Dunn et al., 1997). This method which has the highest sensitivity among noninvasive tests is also used in seroprevalence studies (Dunn et al., 1997; Malaty et al., 2002).

*H. pylori* positivity was determined in 119 patients and *H. pylori* negativity was determined in seven patients by urea breath test in the present study. As compared to the gold standard methods, seven false positive results and four false negative results were obtained. False positive results may be observed as urea breath test is a test based on urease. In our study, the urea breath test was the test with the highest sensitivity (96.5%) among all tests. In the study performed by Lu et al., (2005) on 60 patients who had experienced peptic ulcer hemorrhage, the sensitivity and specificity of urea breath test was reported to be 94% and 85%, respectively. Vaira and Vakil investigated the noninvasive methods and determined the specificity and sensitivity of urea breath test to be 94.7% and 95.7%, respectively. Moreover, in that particular study, the researchers reported that urea breath test had the highest sensitivity and specificity among noninvasive tests and was a suitable method to be used for diagnosis and treatment control (Hegedus et al., 2002). The sensitivity of the urea breath test in our study was similar to that in other studies; however, its specificity was determined to be lower than that in similar studies due to its high false positivity. Urea breath test was considered to be a reliable noninvasive test for the diagnosis and the control of eradication of *H. pylori* but verification test should be performed for suspicious situations.

While the sensitivity of culture method was found to be 68.1% in determining *H. pylori*, the sensitivities of other five methods was found between 85% and 95%. While the specificities were calculated to be 100% for both culture and histopathological examination methods, the specificities of two invasive methods, PCR and rapid
urease test, was determined to be 70%. Among invasive methods, the specificities of ELISA and urea breath test were observed to be low in our study. PPV of all tests was determined to be higher than 94%; the histopathological examination was found to have the highest NPV.

The results of our study showed that the diagnostic value of invasive methods was higher than that of noninvasive methods.

Urea breath test is the most convenient test in the routine practice for scanning and control after medical treatment. The stool antigen tests become an alternative to urea breath test especially in children with increased use. As the serologic tests are not a proof of active infection, they are ideal for seroprevalence studies.

In the present study, no significant difference was found between PCR method and gold standard in terms of the rate of H. pylori positivity. The use of primers targeting 23S rRNA and the use of nested PCR method in our study increased the specificity of PCR. The determination of H. pylori by PCR is on the way to become one of the routine diagnostic methods.

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