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Publication date
1999

Published in
Journal of Clinical Microbiology

JOURNAL OF CLINICAL MICROBIOLOGY, Dec. 1999, p. 4150–4152 Vol. 37, No. 12
0095-1137/99/$04.00+0

Evaluation of Three Commercial Serological Tests with Different Methodologies To Assess *Helicobacter pylori* Infection

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Received 19 April 1999/Returned for modification 21 August 1999/Accepted 7 September 1999

The sera of 142 *Helicobacter pylori*-positive and 32 *H. pylori*-negative patients were assessed by a desktop test (QuickVue), an enzyme-linked immunosorbent assay (ELISA) (HM-CAP), and a solid-phase, two-step chemiluminescent enzyme immunoassay (Immulite). These tests yielded sensitivities of 97, 97, and 91% and specificities of 97, 94, and 100%, respectively. In conclusion, the desktop test and the ELISA are more sensitive than the chemiluminescent enzyme immunoassay ($P < 0.05$). The chemiluminescent enzyme immunoassay has the advantage that it is fully automated.

Helicobacter pylori infection in humans is one of the most widespread infections today, and its cure prevents peptic ulcer recurrence (11). Besides chronic gastritis and peptic ulcer disease, *H. pylori* infection is strongly associated with gastric cancer and cancer of mucosa-associated lymphoid tissue (2, 3, 6) and is diagnosed by culturing of gastric biopsy specimens. Noninvasive tests like the urea breath test and tests based on serology may be an alternative for assessing *H. pylori* infection (1, 4, 8). Serology tests can be based on either the detection of *H. pylori* antigens in the feces of patients or anti-*H. pylori* antibodies in the patients' blood or saliva (8, 9). The exact role of serology in the management of *H. pylori* infection has still to be defined, although there is evidence that, used as a screening procedure, it can reduce endoscopy cost and workload (10, 13). The aim of this study was to evaluate the sensitivity and specificity of three commercially available serology tests, each with a different methodology to assess *H. pylori* infection. The desktop test (QuickVue; Quidel, San Diego, Calif.) is a one-step test that was originally designed to be used as a whole-blood test but can also be used for serum specimens. The results can be read 10 min after the submission of the sample to the test. The HM-CAP test (Enteric Products, Inc., Stony Brook, N.Y.) is a standard enzyme-linked immunosorbent assay (ELISA). Sera are added to wells, precoated with *H. pylori* antigens, in a 96-well microtiter plate. The test is completed, including reading of the results, within 1 h. This ELISA is qualitative and uses calibration sera to convert absorbance to arbitrary values. The Immulite test (Diagnostic Products Corporation, Los Angeles, Calif.) is a solid-phase, two-step chemiluminescent enzyme immunoassay. The solid phase, a polystyrene bead enclosed within a test unit, is coated with partially purified *H. pylori* antigen. The diluted serum sample and a protein-based buffer are simultaneously introduced into the test unit and incubated for approximately 30 min with intermittent agitation. During this time, *H. pylori*-specific immunoglobulin G (IgG) will bind to the antigen-coated bead. Unbound serum is then removed by washing by means of centrifugation. An alkaline phosphatase-labeled anti-human IgG is introduced, and the test unit is incubated for another 30-min cycle. Again, the unbound conjugate is removed by a centrifugal wash. Then the chemiluminescent substrate is added, and the test unit is incubated for a further 10 min. The chemiluminescent substrate, a phosphate ester of adamantyl dioxetane, undergoes hydrolysis in the presence of alkaline phosphatase to yield an unstable intermediate. The continuous production of this intermediate results in the sustained emission of light, thus improving precision by providing a window for multiple readings. The bound complex and thus also the photon output, as measured by the luminometer, are related to the presence of anti-*H. pylori* IgG in the sample. A qualitative result is then obtained by comparing the patient serum result to an established cutoff. This system automatically handles the serum sample and reagent additions, the incubation and separation steps, and measurement of the photon output via the temperature-controlled luminometer. Test results for controls and patient samples are obtained from comparison of the observed signal with a cutoff derived from the adjuster's response and the bar-coded parameters. A printed report is generated after completion of the test.

(Part of this work was presented at the 98th General Meeting of the American Society for Microbiology [10a]).

One hundred forty-two *H. pylori*-infected dyspeptic adults (70 with peptic ulcer disease and 72 with gastritis only) underwent gastroduodenoscopy at the Department of Gastroenterology in the Academic Medical Center, Amsterdam, The Netherlands (12). During each endoscopic procedure, three antral and three corpus mucosal biopsy specimens were obtained by use of sterile biopsy forceps. One antrum and one corpus biopsy specimen were placed in 2 ml of phosphate-buffered

TABLE 1. Sensitivity, specificity, and positive and negative predictive values of three *H. pylori* serology tests[a]

Test used	Sensitivity	Specificity	Positive predictive value	Negative predictive value
Desktop	97 (138/142)	97 (31/32)	99 (138/139)	89 (31/35)
ELISA	97 (138/142)	94 (30/32)	99 (138/140)	88 (30/34)
Two-step solid phase	91 (129/142)	100 (32/32)	100 (129/129)	71 (32/45)

[a] Values are shown as percentages (number of samples correctly identified/total number of samples).

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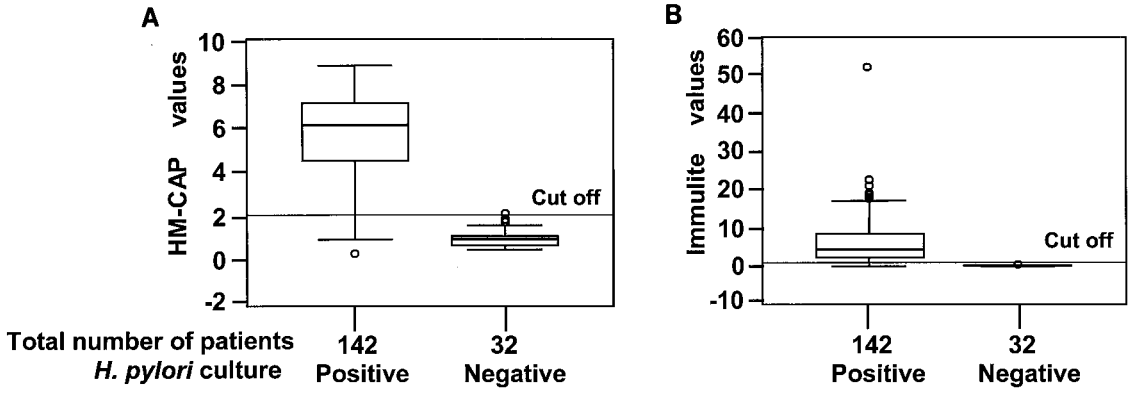

FIG. 1. Summary plot based on the median, quartiles, and extreme HM-CAP (A) and Immulite (B) values, obtained with the sera of 142 *H. pylori*-positive patients and 32 *H. pylori*-negative controls. The box represents the interquartile range which contains the middle 50% of values. The whiskers extend to the highest and lowest values, excluding outliers. The line across the box indicates the median.

saline at 4°C and used for bacteriological culturing. The other four specimens were fixed in 10% formalin for histopathological examination. Cultures were prepared by smearing biopsy specimens on the surface of horse blood agar plats (7% defibrinated horse blood Columbia agar base; Oxoid CM 331; Unipath, Basingstoke, England) and horse blood agar plates containing Skirrow supplement (Unipath). *H. pylori* organisms were identified on the basis of typical colony morphology; characteristic appearance on Gram staining; and positive urease, oxidase, and catalase tests. *H. pylori* infection was present if either culture and histopathological assessment or only histopathology assessment was positive. Controls were 32 noninfected patients. They had *H. pylori*-negative cultures and normal histopathology of multiple gastric biopsy specimens for at least 4 years prior to serological testing. Blood was drawn from the *H. pylori*-positive patients and from the *H. pylori*-negative controls by venous puncture. After clotting, serum was stored at −70°C in small aliquots. Sera were assessed by the three different serology tests according to the manufacturers' protocols. The results obtained with the three different tests with the sera from 142 *H. pylori*-positive patients and 32 *H. pylori*-negative patients are presented in Table 1. The sensitivities of the QuickVue, the HM-CAP, and the Immulite tests were 97, 97, and 91%, respectively (Table 1) ($P < 0.05$). The three tests had specificities of 97, 94, and 100%, respectively (not significant). The positive predictive values of the three tests were 99, 99, and 100%, respectively. The negative predictive values of the tests were 89, 88, and 71%, respectively (differences not significant). The sensitivity values of the ELISA and the desktop test were in the same range as those reported by others (4, 5). The specificity of the tests in this study was somewhat higher. This may be explained by the *H. pylori*-negative controls included in this study. The subjects in this group were *H. pylori* negative by culture and histopathology over a prolonged period of at least 4 years. This significantly diminishes the chance of finding serological false positives in this group, because the concentration of anti-*H. pylori* antibodies, elicited by a possible infection prior to the *H. pylori*-negative period, would be strongly reduced during that *H. pylori*-negative period. The desktop test is designed to be used as a whole-blood test and awaits evaluation as such, especially in a primary care setting. In contrast to the QuickVue test, the HM-CAP and Immulite tests provide quantitative data (Fig. 1). The dynamic range of the Immulite test is wider than that of the HM-CAP test, which may be beneficial for posttreatment evaluation.

In conclusion, the three serology tests are sensitive and specific tests to assess *H. pylori* infection. The desktop test and the ELISA are more sensitive than the chemiluminescent enzyme immunoassay, but the QuickVue desktop test provides only qualitative results. However, the desktop test has the advantage of obtaining results within minutes. The ELISA HM-CAP and the chemiluminescent enzyme immunoassay Immulite are both quantitative, but the latter test has the advantage that sample handling, reading, and interpretation are fully automated. The design of the Immulite test, i.e., accurate quantification by using internal controls and a wide dynamic range in output values, makes it potentially suitable to assess *H. pylori* eradication by comparison of the patient's pretreatment serum with the posttreatment serum.

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