Utility of Stool Sample–based Tests for the Diagnosis of *Helicobacter pylori* Infection in Children

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ABSTRACT

Objective: *Helicobacter pylori* antigen or DNA in stool are meant to detect the bacteria; however, in children the colonization of the gastric mucosa by *H pylori* is usually weak and fecal excretion of antigen or DNA varies considerably, challenging the utility of these tests in this age group. The aim of the present study was to carry out a systematic review and meta-analysis to evaluate the performance of stool *H pylori* DNA and antigen tests for the diagnosis of infection in children.

Methods: We conducted a systematic review and meta-analysis to assess the accuracy of stool tests for diagnosis of *H pylori* infection in children. We searched PubMed, EMBASE, and LILACS databases. Selection criteria included participation of at least 30 children and the use of a criterion standard for *H pylori* diagnosis. In a comprehensive search, we identified 48 studies.

Results: Regarding antigen-detection tests, enzyme-linked immunosorbent assay (ELISA) monoclonal antibodies showed the best performance, with sensitivity and specificity of 97%, positive likelihood ratio (LR+) of 29.9, and negative likelihood ratio (LR−) of 0.03. ELISA polyclonal antibodies had sensitivity of 92%, specificity of 93%, LR+ of 16.2, LR− of 0.09, and high heterogeneity ($P < 0.0001$). One-step monoclonal antibody tests demonstrated sensitivity of 88%, specificity of 93%, LR+ of 10.6, and LR− of 0.11. For DNA detection, polymerase chain reaction–based test showed sensitivity of 80.8%, specificity of 98%, LR+ of 17.1, and LR− of 0.18.

Conclusions: Detection of *H pylori* antigen in stools with ELISA monoclonal antibodies is a noninvasive efficient test for diagnosis of infection in children. One-step tests showed low accuracy and more studies are needed to obtain a useful office-based screening test. The available molecular tests are still unreliable.

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Both invasive (eg, endoscopy with biopsy for histology, culture, and rapid urease test [RUT]) and noninvasive (eg, urea breath test [^{13}C-UBT], stool antigen test) methods are available to diagnose *Helicobacter pylori* infection in children (1–3). Ideally, a diagnostic test for *H pylori* infection in children should be noninvasive, highly sensitive and specific, inexpensive, and easy to perform. Because *H pylori* and/or its macromolecules such as proteins and DNA are shed in feces, stool-based tests such as culture, antigen, or DNA detection have become accepted techniques for noninvasive diagnosis. The use of stool culture for isolation of *H pylori* has limitations because the organism is usually already dead or present in a nonviable coccoid form (4,5). This leaves either stool *H pylori* DNA or antigen-detection tests as more suitable diagnostic tools (6–8).

Several commercial enzyme-linked immunosorbent assay (ELISA) tests for *H pylori* stool antigen are available. The main differences among these tests are the nature of the detecting antibodies; some kits use a polyclonal anti–*H pylori* capture antibody, such as Premier Platinum HpSA ELISA (Meridian Diagnostic, Cincinnati, OH), whereas other assays use monoclonal antibodies such as Amplified IDEIA HpStAR (Dako, Glostrup, Denmark) and Femtolab (Martinsried, Germany). Recently, a new, rapid, 1-step test (immunochromatographic format) using monoclonal antibodies has been introduced, (ImmunoCard STAT! HpSA Meridian Diagnostic, and RAPID Hp StAR, Dako). This test is easy, requires only 5 minutes to complete, and can be performed in a doctor's office (9–13).

Tests that detect *H pylori* DNA are based on polymerase chain reaction (PCR) amplification of target sequences such as *ureA, ureC,* and *16S rRNA* genes (7,14). In vitro studies with reference strains have shown that although coccoid forms are not viable, they contain DNA and RNA material that is readily detected by PCR (5,14,15). However, PCR-based tests depend on the quality and amount of DNA recovered, the target sequences, and the nature of the amplification protocol, such as nested PCR, real-time PCR, and the like. Several population-based studies evaluating the use of PCR-based tests have reported with variable and inconsistent results, with sensitivities ranging from 25% to 100% and specificities ranging from 80% to 100% (16–19).

Given the diversity of stool-based diagnostic tests for *H pylori* infection, a critical appraisal of their performance is needed. Several meta-analyses on the topic have been published based predominantly on adult populations and populations of adults and children combined (6,20,22). One meta-analysis of the performance of the commercial test HpSA including 43 studies

reported sensitivity and specificity values of 92.4% and 91.9%, respectively, using ^{13}C-UBT as the criterion standard (6). Another meta-analysis of HpSA that only included 6 studies enrolling patients with bleeding peptic ulcer reported lower sensitivity (87%) and specificity (70%) (21). Another recent meta-analysis with 22 studies found that the commercial monoclonal anti–*H pylori* capture antibody (IDEIA HpStAR) achieved high sensitivity (94%) and specificity (97%) (22). We are unaware of a meta-analysis of the accuracy of stool-based tests in which studies included only children as participants. An initial search of the literature identified 64 studies evaluating *H pylori* antigen or DNA stool tests in children. Therefore, we carried out a systematic review and meta-analysis to evaluate the performance of stool *H pylori* DNA and antigen tests for the diagnosis of infection in children.

METHODS

We followed standard guidelines and methods for systematic reviews and meta-analysis of diagnosis (23–26).

Search Strategy

We systematically searched PubMed, EMBASE, and LILACS (June 1998–May 2007) to identify studies evaluating the accuracy of stool *H pylori* DNA or antigen tests for the diagnosis of *H pylori* infection in children from birth and up to 19 years old. The search terms used included "*Helicobacter pylori,*" "children," "antigen detection," "diagnostic," "stool samples," "nucleic acid amplification," "polymerase chain reaction," "sensitivity," "specificity," "feces," and "detection." In addition, we checked references of eligible articles and corresponded with authors when a full-length article was not available directly online.

Study Eligibility

We included articles that met the following predetermined criteria: comparison of *H pylori* stool tests with a reference standard (including culture, histological examination, or UBT) (27) and evaluation of a minimum of 30 participants. Although our initial literature search had no language restrictions, studies not available in either English or Spanish were excluded. In addition, we excluded reviews, letters to the editor, opinions, and recommendations about the diagnosis of *H pylori* infection in children.

Outcomes

Our analysis focused on the following measures of diagnostic accuracy: sensitivity (true-positive results), defined as the proportion of tests determined to be correctly identified as *H pylori* positive by a reference standard (culture, histology, or UBT); specificity (true-negative results), defined as the proportion of tests determined to be negative; positive likelihood ratio (LR+), which measures how many times a positive test is more likely found in infected versus noninfected children; negative likelihood ratio (LR−), which measures how many times a negative result is more likely found in infected versus noninfected children; and diagnostic odds ratio (DOR), defined as the ratio of the odds of a positive test result in a child with infection compared with a child without infection. The DOR combines sensitivity and specificity into a single measure of test accuracy, ranging from 0 to infinity, with higher values indicating better discriminatory test performance or higher accuracy (24,28).

Data Extraction

Two reviewers (J.A.S. and J.V.R.) extracted data from all of the included articles using a predefined database in Microsoft Excel format. A second reviewer (L.L.F.) independently extracted data of a subset (20 articles [32%]) of the included articles. The interrater agreement between the 2 reviewers for sensitivity and specificity was 95%; differences were solved by consensus. The following data were extracted: study design; criterion standard for *H pylori* diagnosis; type of test: ELISA and immunochromatography assays for *H pylori* antigen detection and PCR for DNA detection; direction of data collection (prospective or retrospective); number of participants; age; language (English or Spanish); and data sufficient to construct a 2×2 table for true-positives, false-positives, true-negatives, and false-negatives. With this information we calculated for each study the following values: sensitivity, specificity, LR+, LR−, and DOR and their corresponding 95% confidence intervals (95% CI).

Assessment of Study Quality

The quality of the studies was validated using reported tools (25,26) for quality assessment of diagnostic studies: was there a comparison of the *H pylori* stool test with an appropriate reference standard? was the *H pylori* stool test result performed in a blinded manner? did the whole sample or a randomly selected subset of the sample receive verification using the reference standard? and did the study recruit prospectively consecutive children suspected of having *H pylori* infection?

Data Synthesis and Meta-analysis

Meta-analysis was done with a fixed-effects model using MetaDiSc Beta-1.4 software (Universidad Complutense, Madrid, Spain) (29). Forest plots were created to display estimates of sensitivity and specificity and to examine heterogeneity (variability between studies). We summarized the joint distribution of true-positive rate (TPR) and true-negative rate (1 false-positive rate [FPR]) with a summary receiver operating characteristic curve (SROC). The SROC curve in studies of diagnostic accuracy represents the relation between TPR and FPR across studies when test performance is evaluated at varying diagnostic thresholds (30). Each study is a separate unit of analysis and contributes an estimate of TPR and FPR. The overall diagnostic performance of a test can be judged by the position and appearance of the SROC, which is fitted by using a regression model. The area under the curve (AUC) represents an overall summary measure of the curve and the test's overall ability to accurately distinguish cases from noncases. Whereas an AUC of 1 represents perfect discriminatory ability, the Q* index represents a summary of test performance in which sensitivity and specificity are equal. A Q* index of 1.0 indicates perfect accuracy (sensitivity and specificity of 100%). Both AUC and Q* range from 0 to 1, with higher values indicating better test performance (24,30).

Selection of Subgroups for the Meta-analysis and Exploration of Heterogeneity

Heterogeneity in meta-analysis refers to the degree of variability among study results (eg, variability in sensitivity estimates). Heterogeneity may result from differences in threshold (eg, cutoff values), disease spectrum, variation in test protocols, and study quality across studies. When significant heterogeneity is present, summary estimates from meta-analyses are hard to interpret. In this

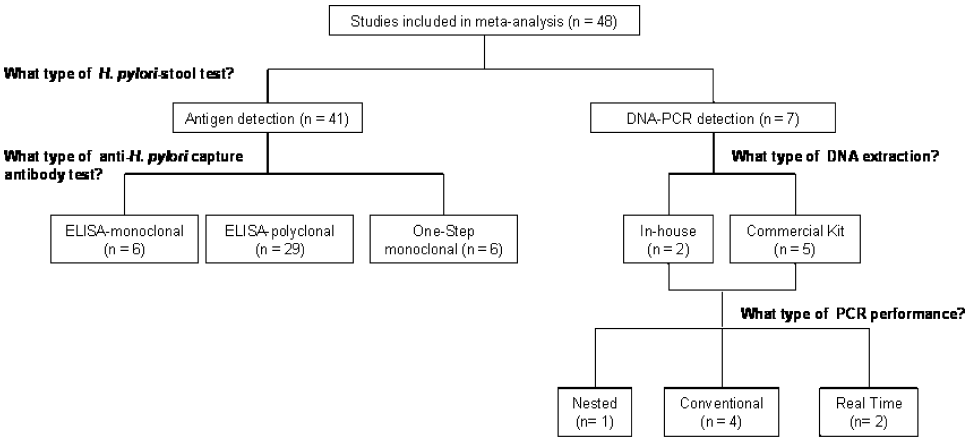

FIGURE 1. Selection of subgroups for exploration of heterogeneity.

meta-analysis we recognized that studies were heterogeneous in several ways, including the type of diagnostic stool test and the different protocols for anti–*H pylori* antigen detection. Therefore, we defined subgroups for the meta-analysis by dividing studies into antigen-detection tests (includes 6 different commercial tests) and DNA-detection tests (includes 6 different target sequences). We further divided the antigen-detection subgroup according to the type of antibody-capture test into the following: ELISA polyclonal, ELISA monoclonal, or 1-step monoclonal (Fig. 1). To explore reasons for heterogeneity, the polyclonal ELISA subgroup was further divided by storage temperature (-20°C vs -70°C); blinded status (at least single-blinded interpretation vs not reported); type of criterion standard (culture/histology vs histology/RUT vs UBT); cutoff value (fixed by ROC curve vs not fixed); and number of filters used for ELISA optical density reading (single vs dual wavelengths vs not reported). Heterogeneity was assessed using the χ^2 test with MetaDiSc Beta-1.4 software (Universidad Complutense) (29).

RESULTS

Study Selection

The literature search identified 405 potentially relevant citations, of which 39 articles (37 English- and 2 Spanish-language articles) were included (Fig. 2) (31,32). Eight (20.5%) of these reported the evaluation of >1 diagnostic test. In these cases, each test comparison was counted as a separate study. Thus, a total of 48 test comparisons (hereafter referred to as studies) were included in the meta-analysis.

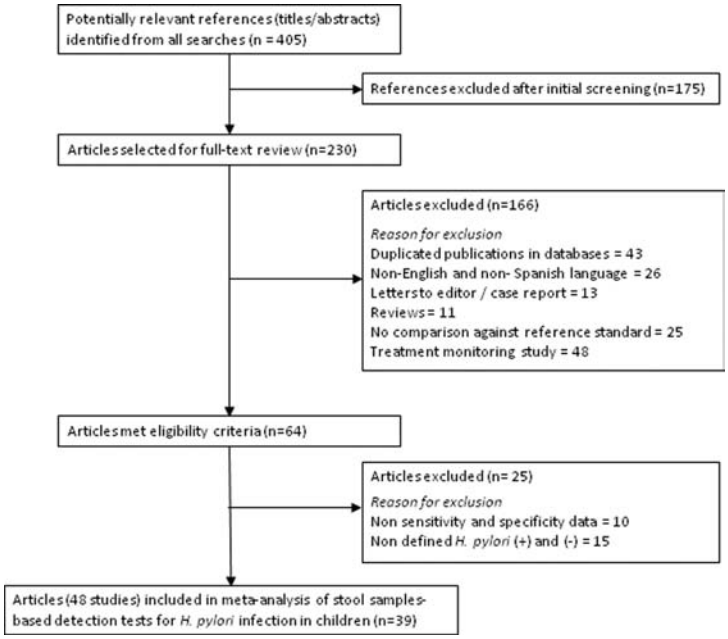

FIGURE 2. Study selection process and reason for exclusion.

TABLE 1. Summary measures of test accuracy for all included studies

Test property (*H pylori* +/−)	Summary measure of test accuracy (95% CI)	Test for heterogeneity P^*
ELISA polyclonal antibody = studies 29 (1393/2460)		
Sensitivity	92.0 (90.0–93.0)	<0.001
Specificity	93.0 (91.0–94.0)	<0.001
LR+	16.2 (10.4–25.2)	<0.001
LR−	0.09 (0.06–0.13)	<0.001
DOR	250.2 (120.4–520.2)	<0.001
ELISA monoclonal antibody = studies 6 (326/445)		
Sensitivity	97.0 (94.0–98.0)	0.83
Specificity	97.0 (95.0–98.0)	0.017
LR+	29.90 (10.3–86.9)	0.006
LR−	0.03 (0.02–0.07)	0.74
DOR	877.7 (230.9–3333.4)	0.05
One-step monoclonal antibody = studies 6 (359/420)		
Sensitivity	88.0 (85.0–92.0)	<0.001
Specificity	93.0 (90.0–95.0)	0.53
LR+	10.60 (7.5–14.8)	0.42
LR−	0.11 (0.05–0.24)	<0.001
DOR	95.51 (38.0–240.1)	0.016
PCR DNA detection† = studies 7 (245/151)		
Sensitivity	80.8 (75.3–85.6)	<0.001
Specificity	98.0 (94.3–99.6)	0.55
LR+	17.1 (7.8–37.3)	0.91
LR−	0.18 (0.08–0.37)	<0.001
DOR	138.4 (47.8–400.5)	0.63

CI = confidence interval; DOR = diagnostic odds ratio; ELISA = enzyme-linked immunosorbent assay; LR− = negative likelihood ratio; LR+ = positive likelihood ratio; PCR = polymerase chain reaction; SSG = species-specific protein antigen.

* χ^2 test for heterogeneity.

† Gen Target: ureA (1), cagA (1), vacA (1), 16sRNA (1), 23sRNA (2), and SSG (1).

Characteristics of Included Articles

The criterion standards reported among the 39 articles varied: 22 (56.4%) used both *H pylori* culture and histology (9,11–13,31,33–49), 5 (12.8%) reported only histology and either RUT or UBT (32,50–53), 8 (20.5%) used UBT alone (10,54–60), and 4 (10.3%) used UBT combined with ELISA (61–64). Blinding status also varied among the articles. Seventeen (43.6%) articles reported at least single-blinded interpretation of results, whereas 22 (56.4%) articles did not state blinding status. One (2.5%) (64) article that assessed the performance of PCR reported the use of a randomly selected subset of the sample for validation with the reference standard. All of the articles reported studies that used a case-control design and prospective data collection. A total of 5799 children were included in the meta-analysis. Of these, 2323 were *H pylori* positive (cases) and 3476 were *H pylori* negative (controls) compared with the respective criterion standard. The median sample size was 107 participants (interquartile range 66–176).

H pylori Antigen Detection

ELISA Polyclonal Antibody

Twenty-nine studies (3853 children) evaluated the accuracy of antigen detection by the ELISA polyclonal anti–*H pylori* capture test. The majority of studies assessed the performance of Premier Platinum HpSA (Meridian Diagnostic) and 1 study, the performance of HpSA-Equipar (Equipar Diagnostici, Saronno, Italy). All summary measures of test accuracy were high (Table 1). Overall accuracy was also high, with an AUC of 0.98 and a Q* of 0.94 (Fig. 3). However, as seen in the corresponding Forest plots (Fig. 4), sensitivity (67%–100%) and specificity (61%–100%) values were variable, suggesting the presence of significant heterogeneity (discussed below).

ELISA Monoclonal Antibody

Six studies (771 children) assessed the performance of commercial ELISA monoclonal anti–*H pylori* capture tests, 4 studies evaluated Amplified IDEIA HpStAR (Dako), and 2 studies evaluated Femtolab. Pooled measures of performance were high (Table 1). Values of AUC (0.99) and Q* (0.96) were also high (Fig. 5A). Although sensitivity estimates were consistently high (93%–98%), the specificity estimates (88%–99%) were variable (Fig. 6). The test of heterogeneity for the DOR was of borderline significance ($P = 0.05$; Table 1).

One-Step Monoclonal Antibody

Six studies (779 children) assessed the performance of immunochromatography tests: 5 studies with the ImmunoCard STAT! HpSA and 1 with the RAPID Hp StAR. Pooled accuracy

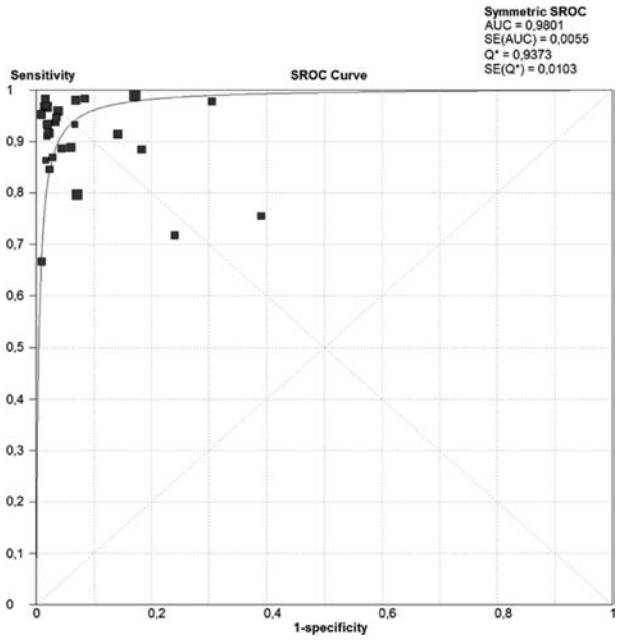

FIGURE 3. SROC for ELISA polyclonal antibody diagnostic tests. Each solid square represents an individual study in the meta-analysis. The curve is the regression line that summarizes the overall diagnostic accuracy. AUC = area under curve; Q* = index defined by point of the SROC in which the sensitivity and specificity are equal; SE (Q*) = standard error of Q* index; SE (AUC) = standard error of AUC; SROC = summary receiver operator curve.

estimates showed moderate performance (Table 1). The AUC of 0.97 and Q* of 0.92 indicated high overall accuracy (Fig. 5B). The DOR was 95.5 (95% CI 38–240.1) and the corresponding test for heterogeneity was significant ($P = 0.016$).

H pylori DNA Detection

The pooled accuracy of nucleic acid detection tests was assessed in 7 studies (396 children). Three of the 7 studies used a single-reaction PCR protocol; of these, 2 studies used immunomagnetic-bead separation (57,61), 1 study used real-time PCR (43), 2 studies used radiolabeled oligonucleotides (64), and the last study used a seminested-PCR test (63). The amplified target genes were as follows: 16sRNA (1), 23sRNA (2), ureA (1), vacA (1), cagA (1), and 1 test based on the DNA sequence of a species-specific protein antigen (Table 1). Different methods for nucleic acid extraction were used: 2 (28.5%) studies used in-house techniques and 5 (71.5%) studies used commercial kits. As seen in Table 1, specificity estimates were high (93%–100%), whereas sensitivity estimates were variable (54%–100%). The overall accuracy of this group was high, with an AUC of 0.98 and a Q* of 0.94 (Fig. 7). The pooled DOR was 138.4 (95% CI 47.8–400.6), and the corresponding test for heterogeneity was not significant ($P = 0.63$; Table 1).

Exploration of Heterogeneity

We explored 5 possible causes for heterogeneity: storage temperature, blinded status, type of criterion standard, diagnostic cutoff value, and the use of single- versus dual-wavelength filters among the subgroup of studies using ELISA polyclonal tests. As seen in Table 2, significant heterogeneity was present for storage temperature, blinded studies, and diagnostic cutoff variables, whereas for type of criterion standard and wavelength number, there was no significant heterogeneity. The pooled accuracy parameters in the subgroup of studies using ELISA polyclonal tests should therefore be interpreted with caution.

DISCUSSION

Principal Findings

This systematic review identified 48 studies that addressed the diagnostic accuracy of stool sample tests for *H pylori* infection in children. The results of the meta-analysis showed that ELISA monoclonal antibody tests (6 studies) have high pooled sensitivity (97%, 95% CI 94–98) and specificity (97%, 95% CI 95–98) with a significant test for heterogeneity ($P = 0.05$); although ELISA polyclonal antibody tests (29 studies) yielded high pooled sensitivity (92%, 95% CI 90–93) and specificity (93%, 95% CI 91–94), there was considerable variation in results; 1-step monoclonal antibody tests were less accurate than either polyclonal or monoclonal ELISA tests; in addition, heterogeneity among these studies was significant ($P = 0.016$), and DNA PCR-based tests yielded variable sensitivities (54%–100%) and high specificities (93%–100%). Because our meta-analysis showed considerable evidence of heterogeneity among studies, we recommend that the results be interpreted with caution.

Antigen-detection Tests

Although there were only 6 studies using the ELISA monoclonal test in children, they reported consistently similar high estimates of sensitivity and specificity (97%). Also, the LRs had good performance: LR+ of 29.9 and LR− of 0.03, meaning that children with *H pylori* infection have a 29.9-fold greater chance of being positive than children without the infection. This is a good ratio to guide the clinician to rule in (ie, confirm) infection. Similarly, an LR− of 0.03 is sufficiently low to confidently exclude *H pylori* infection when an ELISA monoclonal test result is negative (65,66). Thus, this ELISA test allows an accurate diagnosis in the absence of annoying and potentially harmful invasive procedures in children, and it is a convenient assay to test *H pylori* infection in the absence of specific symptoms or even in community-based surveys.

Overall, the ELISA monoclonal antibody tests perform better than the ELISA polyclonal antibody tests, probably because of a higher affinity and specificity of the antibodies; still, specificity values for both tests showed considerable heterogeneity. The possible reasons for this heterogeneity include variability in the study settings, reference standards, and differences in age, genetics, and clinical spectrum of the patients studied (67–69). For example, Frenck et al (37) determined that in children younger than 6 years, the specificity of the ELISA monoclonal antibody test was 81%, whereas in children older than 6 years, specificity increased to 100%.

Of the 3 available *H pylori*–antigen-detection tests, the 1-step version of the monoclonal antibody test showed the lowest accuracy, a low estimated sensitivity (88%), high heterogeneity, and high LR− of 0.11. With these values, the infection cannot be excluded when test results are negative. An important drawback with the 1-step tests is that they rely on individual visual interpretation of the results, and the interobserver variability and equivocal results are of concern, especially when the test is weakly positive (3,13). In its current form, this test is unreliable and its use is not recommended in children.

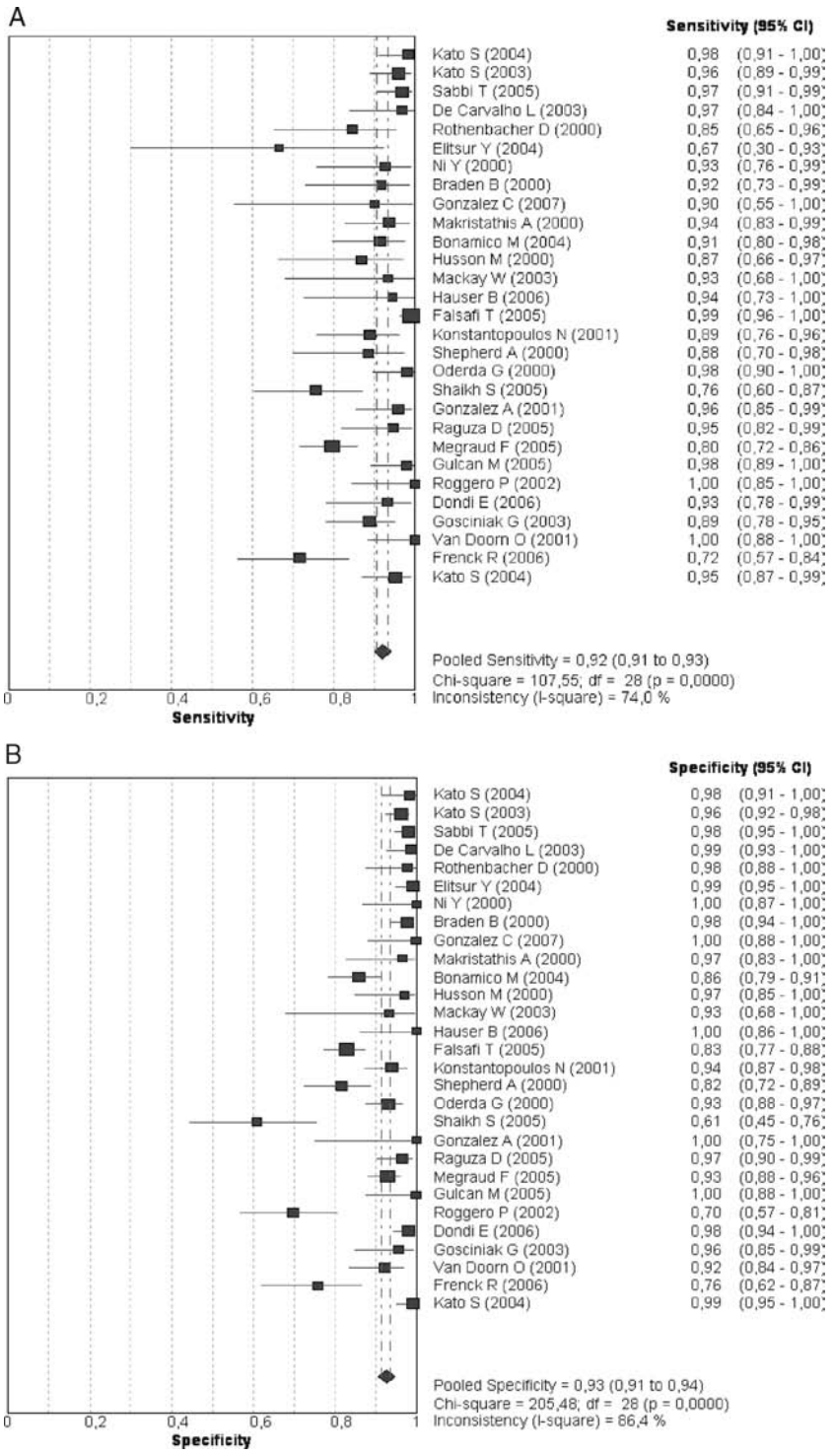

FIGURE 4. Forest plot for the sensitivity and specificity of the enzyme-linked immunosorbent assay polyclonal antibody tests. Squares and lines represent the point estimates and 95% confidence intervals, respectively. The size of the square indicates the study size. The pooled estimated is denoted by a diamond at the bottom.

DNA-detection Tests

Few studies have addressed the use of PCR amplification for the detection of *H pylori* in stool samples with the majority being in-house PCR protocols. Our analyses demonstrated that the *H pylori* DNA PCR detection in stool samples has high specificity (98%) and an acceptable LR+ value (17.1), although sensitivity was low (80.8%) and LR− was high (0.18). One potential explanation for

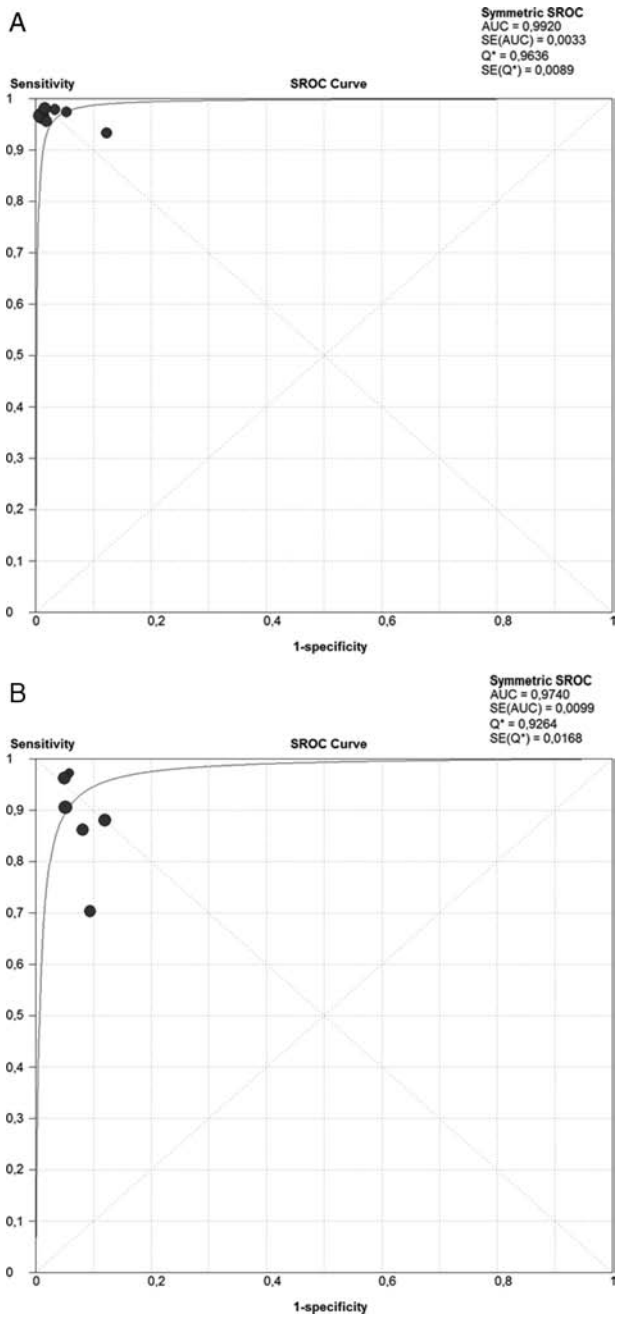

FIGURE 5. SROCs for enzyme-linked immunosorbent assay and 1-step monoclonal antibody tests. Each solid square represents an individual study in the meta-analysis. The curve is the regression line that summarizes the overall diagnostic accuracy. AUC = area under the curve; Q* = index defined by point of the SROC in which the sensitivity and specificity are equal; SE(Q*) = standard error of Q* index; SE(AUC) = standard error of AUC; SROC = summary receiver operator curve.

the lower sensitivity in this subgroup is that PCR is susceptible to inhibitor compounds commonly found in feces (70–72). Some authors have reported the use of gene-capture methods to yield more pure *H pylori* DNA to overcome this problem (57,61,73,74). Another explanation is the increased presence of *H pylori* coccoid forms from which DNA is more difficult to detect by PCR than from rod-shaped cells (14). To improve the accuracy of the DNA detection tests, we recommend performing nested PCR (17,75,76). In addition, PCR requires specialized laboratory facilities and trained personnel, which are usually not available in diagnostic laboratories in developing countries. Still, an important advantage of this test is the potential to genotype virulence genes and to detect infection with antibiotic-resistance strains in children in whom treatment has failed (43,62,64). Recently, a novel commercial real-time PCR kit (ClariRes, Ingenetix, Vienna, Austria) to detect clarithromycin susceptibility of *H pylori* in stool specimens was launched. We analyzed 1 study with this assay, in which the reported sensitivity was low (63%) (43). Further studies are needed to evaluate this test in children.

Exploration of Heterogeneity

The shape of the SROC curve (Fig. 3) of the ELISA polyclonal antibody tests suggests that variability in the thresholds could partly explain the heterogeneity (24,30). An additional variable behind this heterogeneity was storage conditions; samples stored at −20°C instead of −70°C improved the accuracy, giving a DOR estimate value 6.2 times greater than before. Inadequate or repeated thawing events may also explain the poor results obtained in some studies. Using UBT as a criterion standard increased sensitivity and specificity to nearly 100% (Table 2); the DOR value was 3.7-fold higher than when *H pylori* culture/histological examination was used as reference and 4.1-fold higher when the criterion standard was histological examination. These results indicate that UBT makes an appropriate noninvasive reference standard in children (27,77). An appropriate cutoff point represents a crucial factor for the accuracy of the test; no uniformity regarding the cutoff point for positive or negative results was observed across studies included in this meta-analysis. Still, some authors fixed the cutoff value by ROC curve; thus, Bonamico et al (34) reported that 0.160 was an adequate fixed cutoff value in their child population. We analyzed the accuracy of the test before and after fixing the cutoff value, and found that the DOR estimate was about 2 times greater with a fixed cutoff value. This may suggest the need to adjust the cutoff value to the specific studied population when standardizing the protocols. The antigenic differences among the *H pylori* strains present in the stool of children from different populations may also modify the response to the test. Furthermore, our meta-analysis found that results measured with a dual wavelength showed higher accuracy with a DOR estimate 2.4 times greater than a single wavelength (Table 2). Lastly, a low concentration of *H pylori* in the stool samples found in infants and younger children may be below the sensitivity of stool-based tests (78).

Although we controlled for different sources of heterogeneity in the studies that used polyclonal antibodies, some of the variation persisted. This may be explained by variability among the different lots of antibodies, to antigenic difference among the *H pylori* strains used for antiserum production, or to immunological cross-reaction with other *Helicobacter* species such as *H heilmannii, H bilis, H pullorum, H hepaticus*, *or H felis* (79,80). In addition, because the excretion of *H pylori* antigens in feces is not continuous, test results may vary (81,82).

Clinical and Epidemiological Implications

Based on the results of this meta-analysis, clinicians should consider the detection of antigens in feces using ELISA tests, particularly with monoclonal antibodies, to diagnose *H pylori*

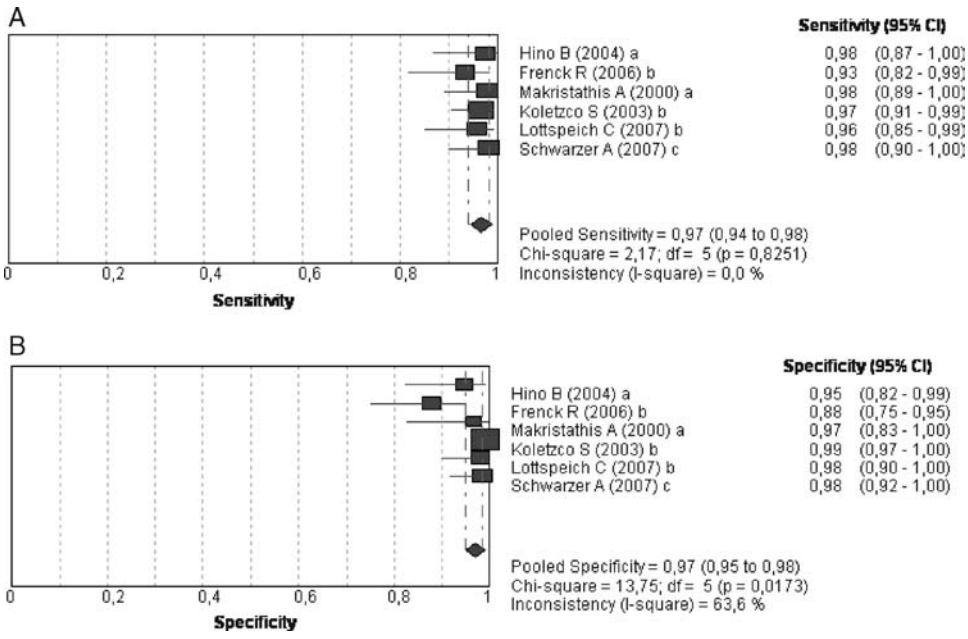

FIGURE 6. Forest plot for the sensitivity and specificity of the enzyme-linked immunosorbent assay monoclonal antibody tests. The squares and lines represent the point estimates and 95% CIs, respectively. The size of square indicates the study size. The pooled estimated is denoted by a diamond at the bottom. CI = confidence interval.

infection in children, especially in young infants because it avoids the need for invasive painful procedures, such as the use of needle to draw blood, or the active collaboration of the child, such as for ^{13}C-UBT. In addition, the test is a noninvasive, simple, rapid test that does not require specialized equipment and is possible to perform at any medical facility. Until recently, treatment decisions in children with severe dyspepsia or chronic abdominal pain were based on the current recommendation by the European Task Force for *H pylori* Infection in Children (2) and the North American Society for Pediatric Gastroenterology, Hepatology, and Nutrition (83), which is to screen symptomatic children with endoscopy because this test allows for the differential diagnosis of abdominal pain including esophagitis, peptic ulcer disease, gastritis, and *H pylori* infection. Thus, the decision to treat children should include first symptoms, endoscopy findings, and then diagnosis for *H pylori* infection. Until recently, it was recommended that ^{13}C-UBT and invasive histological examination or culture from biopsies be used because neither antigen nor DNA detection in feces appears sufficiently reliable for clinical use.

H pylori antigen detection tests may be useful for monitoring the success of eradication therapy; furthermore, because stool samples are easily obtained and can be stored frozen, the test is also suitable for epidemiological studies. To achieve better performance with these tests, it is important to carefully evaluate temperature, cutoff value, or wavelength of the assay, and one should also consider that some reports suggest that antimicrobial agents, proton pump inhibitors, and bismuth preparations reduce the sensitivity of these tests (84).

Strengths and Weaknesses of the Review

An important strength of our study is its comprehensive search strategy. Screening, study selection, and quality assessment

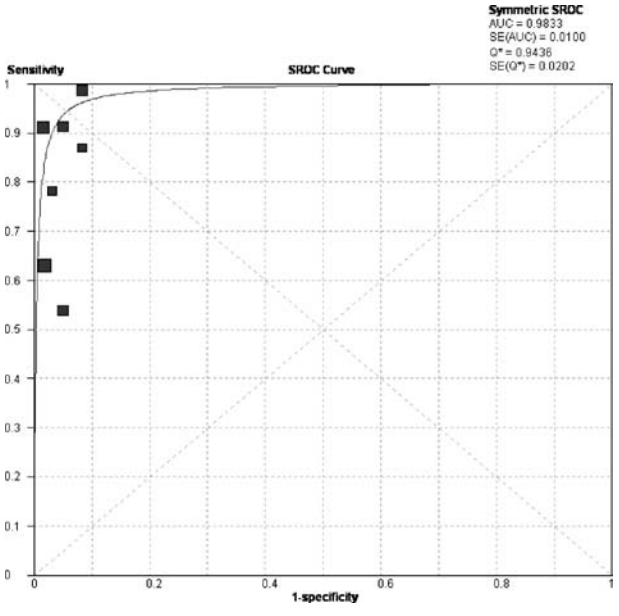

FIGURE 7. SROC for DNA polymerase chain reaction–based tests. Each solid square represents an individual study in the meta-analysis. The curve is the regression line that summarizes the overall diagnostic accuracy. AUC = area under curve; Q* = index defined by point of the SROC in which the sensitivity and specificity are equal; SE(AUC) = standard error of AUC; SE(Q*) = standard error of Q* index; SROC = summary receiver operator curve.

TABLE 2. DOR estimates for ELISA polyclonal test

Study characteristics (n)	DOR (95% CI)	χ^2 test of heterogeneity	*P* for heterogeneity
Storage temperature			
−20°C (25)	322.0 (150.5–688.5)	106.5	<0.0001
−70°C (4)	51.4 (8.9–298.2)	13.9	0.003
Blinding			
Not reported (18)	282.9 (104.0–769.9)	87.0	<0.0001
Blinded (11)	208.4 (67.5–643.4)	52.6	<0.0001
Reference standard			
Culture/histology (16)	224.8 (94.4–535.4)	64.8	<0.0001
Histology/others (4)	199.5 (38.7–1029.0)	61.7	<0.0001
UBT (9)	830.4 (259.9–2652.9)	2.3	0.509
Cutoff*			
Without fixed (9)	266.8 (97.3–731.6)	15.7	0.047
Fixed by ROC curve (9)	435.2 (91.5–2068.8)	28.0	<0.0001
Wavelength measurement			
Single (450 nm) (16)	191.4 (71.3–514.0)	66.2	<0.0001
Dual (450/630 nm) (10)	463.2 (225.1–953.3)	9.1	0.331

CI = confidence interval; DOR = diagnostic odds ratio; ELISA = enzyme-linked immunosorbent assay; ROC = receiver operating characteristic.
* Analysis in only 9 studies with information (true-positive, false-positive, true-negative, false-negative) before and after fixed cutoff.

were done independently by 2 reviewers. For some studies, we reduced the problem of missing data by directly contacting the authors. We also explored heterogeneity and potential publication bias in accordance with published guidelines (24,68,69), and we analyzed data within specific subgroups to lessen the effect of heterogeneity.

We acknowledge that the present study has some limitations; first, we were able to include only English- and Spanish-language articles, and this may have introduced selection bias to our results. Second, we did not address the effect of factors such as laboratory infrastructure, expertise with the technology, and patient demographics. Although we used guidelines such as Standards for Reporting of Accuracy (26) to improve the quality of the analysis, our findings should be interpreted in the context of the quality and variability of the included studies. Unfortunately, we were not able to evaluate the performance of *H pylori* antigen- or DNA-detection tests in feces among different age groups because this information was not available in the majority of the included studies. The latter issue is relevant because diagnostics test results may vary between young children and infants (11,33,36,44,47).

CONCLUSIONS

The results provided by this systematic review and meta-analysis suggest that detection of *H pylori* antigens in stool samples with monoclonal antibody–based ELISA test is a noninvasive and efficient test for the diagnosis of *H pylori* infection in children. The performance of the ELISA test with polyclonal antibodies is inadequate for use in clinical diagnosis and additional adjustments are required. The 1-step polyclonal office-based test showed low accuracy and the protocol should be improved substantially before clinical use. Tests developed to detect DNA are not yet reliable for the diagnosis of *H pylori* infection in children; still, they may be useful to complement studies of virulence genes and resistance to antibiotics.

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