Research Article

COMPARISON BETWEEN UREA BREATH TEST AND STOOL ANTIGEN TEST FOR DIAGNOSIS OF HELICOBACTER PYLORI IN CHILDREN AND ADOLESCENT IN ERBIL CITY

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Abstract

Background
Helicobacter pylori infection, a global health concern, has high childhood prevalence, often starting early and persisting for decades before causing diseases. Diagnostic tests include noninvasive (stool antigen test, urea breath test, blood test) and invasive (histology, rapid urease test, microbiological culture) methods. This study aimed to compare stool antigen and urea breath tests for diagnosing *Helicobacter pylori* in children in Erbil, Kurdistan, and explore variations by age, gender, family size, and parental economic status.

Patients and methods This cross-sectional study, conducted in Erbil, Kurdistan region of Iraq from 1st January 2024 to1st December 2024, involved 49 patients, aged 6 to 16 years. The diagnosis of H. pylori was established through a combining clinical assessments, stool antigen tests or urea breath tests followed by histological examinations after upper gastrointestinal endoscopy. Specificity and sensitivity of both stool antigen test and urea breath test determined by comparing to endoscopic findings.

Results
This study found diagnostic tests, the stool antigen test (HpSA) demonstrated a sensitivity of 73.1%, specificity of 82.6%, and accuracy of 78%. In contrast, the 13C-urea breath test (UBT) showed higher performance metrics with a sensitivity of 88.5% and specificity of 91.3%. Also, strong links between *Helicobacter pylori* infection in children and family history of *Helicobacter pylori* infection. Hematological profiles showed no significant differences. Key symptoms were abdominal pain and vomiting.

Conclusion
The 13C-urea breath test demonstrated high accuracy for H. pylori diagnosis in children.

Key words: Gastritis, Helicobacter pylori, Urea breath test

Introduction

Roughly half of the population is infected by *H. Pylori*, is contracted in childhood, tends to endure in the stomach unless treated with antibiotics, and the risk of infection is associated with factors such as overcrowding, inadequate sanitation, and lower economic status, displaying an inverse correlation with socioeconomic status (1) *H. Pylori* infection in children can manifest with signs like stomach ache, acid indigestion, queasiness, throwing up, and, on rarer occasions, it may result in issues like stubborn iron-deficiency anemia or impaired growth (2).

A variety of diagnostic methods are available for identifying H. Pylori infections, with classifications into non-invasive and invasive approaches. Non-invasive methods include the urea breath test, fecal antigens, serological tests, and molecular methods for H. pylori infection detection, while invasive methods involve endoscopy, histology, and culture (3).

The stool antigen test (SAT) is a direct method for detecting H. pylori infection, relying on the identification of H. pylori antigens in feces (4). Utilizing either enzyme immunoassay or immunochromatography assay methods with monoclonal antibodies, SATs demonstrate superior results compared to polyclonal-based tests (5)

The urea breath test detects converted urea using a radioactive carbon isotope, with patients drinking urea and the resulting carbon dioxide being detectable in the lungs (6). Carbon isotopes ^{13}C and ^{14}C are commonly used, with ^{13}C preferred for pediatric and pregnant patients due to lower radiation exposure (7).

Pediatric *H. Pylori* treatment guidelines differ from those for adults, as emphasized by recent ESPGHAN and NASPGHAN guidelines, which advise against "test and treat" strategies, emphasizing the importance of investigating underlying causes of gastrointestinal symptoms rather than focusing solely on *H. Pylori* presence in children. (8)

The objective of this study was to comparison between stool antigen and urea breath tests in diagnosing *H. Pylori* among children in Erbil, Kurdistan region of Iraq. Also assess their association with certain factors such as age, gender, number of family members, and the economic status of their parents.

Patients and methods

This cross-sectional study was conducted at the Pediatric Gastrointestinal Department of Raparin Hospital, located in the Kurdistan region of Iraq (Erbil), from1st Jan. 2024 to1st Dec. 2024.. Raparin pediatric teaching hospital is the only pediatric governmental hospital in Erbil city- Iraq, located in north-east of Erbil city, it has outpatient clinic and subspecialty outpatients.

This study involved 49 patients, aged 6 to 16 years. The diagnosis of H. pylori was established through a comprehensive approach, combining clinical assessments, stool antigen tests, urea breath tests and histological examinations after upper gastrointestinal endoscopy. Patients referred to endoscopy department with different significant gastrointestinal manifestations and arranged for upper GIT endoscopy.

Exclusion criteria

1.Age >16yrs or <6yrs

2. patient who had taken antibiotic within the last 4wk and PPI taken within 2wks,H2- Blocker within 2 days

3. patient with complicated underlying condition or disease.

Following the acquisition of verbal consent from the family, data were gathered using a prepared questionnaire, and the patient was subsequently sent for laboratory investigation. After thorough history and physical examination, Laboratory investigations included comprehensive blood count and biochemical measurements encompassed WBC (white blood cell count), HB (hemoglobin), PLT (platelet count), IRON, and Ferritin. Specificity and sensitivity of both stool antigen test and urea breath test determined by comparing to (Gold Standard ,gastric biopsy).

Stool samples were promptly processed using stool antigen cards from (Green Lab MEDIKAL, Turkey),

The ^{13}C urea breath examination employing the UREA 13C breath test *H. Pylori* diagnosis kit from Feeel full, China. Following the manufacturer's instructions, a baseline breath sample was collected, and the patient then ingested a urea-containing reagent. After 30 minutes, a second breath sample was taken.

A gastroenterologist conducted diagnostic upper GI endoscopy, during which three biopsies were taken from the antral region following mucosal sampling, one preserved for rapid urease test. Additional biopsies from at least two topographic sites (from both the antrum and the corpus, at the lesser and greater curvature of each) should be taken and clearly labelled in two separate vials. Additional biopsies of visible suspicious lesions should be taken. (8) Cases were categorized into those with positive histopathological evidence of H.Pylori infection (GROUP A) and negative findings (GROUP B).

Results

Among 49 symptomatic children included in the study ,26 of them were endoscopically confirmed H.Pylori infected (Group A) and remaining 23 were negatives(Group B). Table 1 provides a comprehensive overview of selected parameters within the study population, gender distribution shows a non- significant trend, with 61.5% females among positive cases and 65.2% males among negative cases (P=0.062). Age ,gestational age and BMI differences are not statistically significant (P=0.363 and P=0.501, respectively).

Notably, a significant association emerges with family history of infection (P=0.000), as 80.8% of confirmed cases have a positive family history, while only 26.1% of negative cases report such a history. Also ,larger proportion

of infected children significantly associated with lower SES (socioeconomic status) and higher crowding index. These findings highlight potential associations, underscoring the need for further research with larger sample sizes to validate and refine these observations.

There are no significant differences in white blood cell count (WBC), hemoglobin (HB), platelet count (PLT), iron levels, or ferritin levels between the two groups (P>0.05 for all). These findings suggest similar hematological profiles in those children (table 2)

Table 3 provides a snapshot of the outcomes from the 13C-urea breath test (UBT) for detecting *H. Pylori*, contrasting positive and negative results with endoscopy findings. Notably, the table indicates the number and percentage of positive and negative results for both the UBT and endoscopy. Among those with negative biopsy results ,four out of 23 were positive for HpSA and only 2 positive for Urea breath test.

The evaluation of the HpSA test in comparison to Endoscopy reveals notable performance metrics. The fecal test demonstrates a sensitivity of 73.1%, specificity of 82.6%, positive predictive value (PPV) of 82.6%, negative predictive value (NPV) of 73.1%, and an overall accuracy of 78%. These results are indicative of the HpSA and UBT tests efficacy in detecting *Helicobacter pylori* infection. The receiver operating characteristic (ROC) further supports the effectiveness of the 13C-urea breath test in this diagnostic context, with a value of 0.899, as shown in Table 4.

Table 1: Demographic data among studied sample

Variables	GROUP A N=26 (%)	GROUP B N=23 (%)	P-value
Gender			0.062
Male	10(38.5)	15 (65.2)	
Female	16 (61.5)	8 (34.8)	
Age in years (mean ±SD)	9.38 ± 3.15	10.17 ± 2.82	0.363
BMI	19.43 ± 3.20	18.81 ± 3.73	0.501
GA			0.241
Term	25 (96.2)	20 (87)	
Preterm	1 (3.8)	3 (13)	
Family history of H. Pylori inf.	21 (80.8)	6 (26.1)	< 0.001
Crowding index	3.61 ± 1.09	2.82 ± 1.07	0.014
SES (socioeconomic status)			0.048
Upper	4 (15.3)	2 (8.7)	
Middle	4 (15.4)	11 (47.8)	
Lower	18 (69.2)	10 (43.5)	

Table 2: Hematological parameters in studied samples

Variables	GROUP A N=26 (%)	GROUP B N=23 (%)	P-value
WBC (c/mm3)	5281.6 ± 713	5274.7 ± 665.4	0.972
HB (g/dL)	12.5 ± 1.3	12.1± 0.9	0.251

PLT (µL)	323021.5 ± 857113.5	336407.2 ± 73838.9	0.563
IRON (µg/dl)	77.7 ± 21.2	79.6 ± 26.8	0.783
Ferritin (ng/mL)	64.8 ± 50.5	68.5 ± 51.1	0.805

Table 3: The Outcomes of the Stool Antigen Test and Urea Breath test in comparison with the reference standard diagnosis of Endoscopy

	Gold standard endoscopy	
	GROUP A, N=26 (%)	GROUP B, N=23 (%)
Fecal HpSA		
Positive (n, %)	19 (73.1)	4 (17.4)
Negative (n ,%)	7 (26.9)	19 (82.6)
Urea Breath Test		
Positive (n, %)	23 (88.5)	2 (8.7)
Negative (n ,%)	3 (11.5)	21 (91.3)

Table 4: Evaluation of HpSA test and 13C-urea breath (UBT) test in comparison to Endoscopy performance

Tests	Sensitivity %	Specificity %	PPV%	NPV%	Accuracy %	ROC
HpSA	73.1	82.6	82.6	73.1	78	0.778
UBT	88.5	91.3	92	87.5	90	0.899

Discussion:

According to the findings of the current research, the prevalence of *H. Pylori* among symptomatic children (49 participants) was determined to be 53.1% by histopathological diagnosis. A study conducted in Iraq found that the prevalence of H. pylori among Iraqi patients varied between 47.8% and 70.4% across different detection methods. The positive detection rates for each method were as follows: qPCR 81 (70.4%), UBT 79 (68.7%), SAT 77 (67%), RUT 76 (66.1%), Cag-IgG 61 (53%), and culture 55 (47.8%) (9). In contrast, two studies reported a lowest rate of 36% and 31% among children attending school or kindergarten (10 ,11). Moreover, the rate identified in our study fell within the range reported in nearby nations like Iran (between 47% and 64%) (12) and Saudi Arabia (50% of studied 303 sample were infected) (13). A review article concluded that the majority of studies conducted in America indicated an overall prevalence of approximately 50% for *H. Pylori* infection, and *Helicobacter pylori* is commonly acquired during childhood (14). Variations in infection rates could stem from differences in living standards, hygiene practices, and other factors across distinct regions and selected sample criteria.

Various factors contribute to the risk of *H. Pylori* infection in childhood. According to the results of this study, there were no statistically significant differences in gender distribution, BMI and age between positive and negative cases among children. This discovery aligns with earlier findings, which noted the absence of significant associations between age, gender, BMI and *H. Pylori* positivity (15). The published study has demonstrated that there is no disparity in the *H. Pylori* infection rate between girls and boys as reported by many studies (16, 17)

Socioeconomic factors may play a role in influencing the likelihood of acquiring an *H. Pylori* infection (18). In this study, a substantial association (p=0.000) was noted in family history, with 80.8% of positive cases having a positive family history. Notably, the highest rate of *H. Pylori* infection recorded among upper lower and lower socioeconomic status class (34.6 %). In Nigeria, a study reported an *H. Pylori* infection prevalence of 14.3% in

children, with rates of 50% in the upper and lower social classes, respectively. Children from the lower social class were more likely to be infected with *H. Pylori* than those in the upper class, as socioeconomic status showed a significant statistical relationship (P = 0.001) with *H. Pylori* seropositivity (19).

No notable distinctions were observed in white blood cell count (WBC), hemoglobin (Hb), platelet count (PLT), iron levels, or ferritin levels between the two groups (P>0.05 for all). These results indicate comparable hematological profiles in children, whether infected or not with *H. Pylori*, in this study. A study documented that individuals in the *H. Pylori* positive group showed significantly reduced levels of ferritin and vitamin B12 (20). Another study affirmed that there was no significant difference in mean platelet volume between the groups (21).

Endoscopic procedures, the current gold standard for assessing peptic ulcer disease and *H. Pylori* presence, pose more risks and discomfort compared to non-invasive tests (22). The UBT (Urea Breath Test) is a dependable and noninvasive method extensively employed for detecting *H. Pylori* infection in adults but its precision in identifying *H. Pylori* infection in infants and young children is limited (22). There are limited reports on the utilization of the ^{14}C-labeled UBT in children, primarily due to radiation concerns. Moreover, false-negative outcomes may arise in patients recently treated with bismuth compounds, antibiotics, or gastric acid antisecretory agents. A study indicated strong concordance between the outcomes of the ^{13}C Urea Breath Test (UBT) and the stool antigen test for infants and toddlers in both high- and moderate-prevalence regions of *H. Pylori* in South America (23). In this study, the HpSA test showed a sensitivity of 73.1%, specificity of 82.6% and overall accuracy of 78% while The 13C-urea breath test had a sensitivity of 88.5%, specificity of 91.3% and overall accuracy of 90%, supported by an ROC curve value of 0.899. In a cross-sectional study, the effectiveness of the Carbon-13 Urea Breath Test (UBT) for predicting *H. Pylori* infection in patients was assessed, revealing a sensitivity of 76.2% and a specificity of 69.2% (24). A study conducted in Iraq unveiled that the 14C-UBT demonstrated superior performance, boasting a sensitivity of 97.5%, specificity of 97%, and an impressive overall accuracy rate of 97.3% (9). Another study's analysis reveals that 13C-UBT exhibits superior diagnostic precision compared to 14C-UBT, with higher sensitivity (96.60% vs. 96.15%), specificity (96.93% vs. 89.84%), likelihood ratios (LR+ 22.00 vs. 10.10; LR- 0.05 vs. 0.06), and area under the curve (AUC; 0.979 vs. 0.968) (25). A Research indicates that stool enzyme-linked immunosorbent assay (ELISA) using monoclonal antibodies is an effective non-invasive test for diagnosing H. pylori in children, with a sensitivity and specificity of 94% and 97%, respectively (26). Despite these minor differences compared to our results ,both tests especially UBT showed relatively high positive prediction of H pylori infection among children.

Conclusion

The 13C-urea breath test demonstrated high sensitivity (88.5%), specificity (91.3%), and accuracy (90%), indicating its efficacy for diagnosing *H. Pylori* in children. This study identified significant associations between *Helicobacter pylori* infection in children and feeding practices, as well as family history. Hematological profiles showed no significant differences between positive and negative cases.

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