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Comparison of Non-Invasive Tests With Endoscopic Biopsy in Diagnosis of Helicobacter Pylori Infection in Pediatric Patients

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ABSTRACT

Background & Objective: *Helicobacter Pylori is a recognized pathogen for gastritis, acid peptic disease and gastric adenocarcinomas. Usually acquisition of infection occurs in early childhood. The objective of this study is to comparison invasive tests for diagnosis of H. Pylori infection in comparison with the gold standard test (endoscopic biopsy and histopathology).* Method: *We have enrolled 100 Pediatric patients presenting with gastro-intestinal symptoms in our study. Endoscopic biopsy with histopathological examination (H & E Staining), stool antigen and serum immunoglobulin G for H. Pylori (non-invasive tests) were performed in all enrolled cases. Sensitivity, specificity, positive predictive value and negative predictive value of these two non-invasive tests separately and in combination (in series) were compared to the gold standard test.* Results: *23% patients were positive for H. Pylori infection on histopathology (H & E staining) of biopsy specimen. Stool antigen for H. Pylori had a sensitivity of 91.30% & specificity of 98.70%, a positive predictive value of 95.45% and a negative predictive value of 97.43%. Serum immunoglobulin G H. Pylori was 73.91% sensitive, 97.40% specific and had a positive predictive value of 89.47% and a negative predictive value of 92.59%. Combination (in series) of non-invasive tests revealed a sensitivity of 69.56%, a specificity of 100%, a positive predictive value of 100% and a negative predictive value of 91.66%.* Conclusion: *Our study has demonstrated that combination of non-invasive test was as efficacious as the gold standard test. Stool antigen for H. Pylori is a simple, suitable and easily applicable in paediatric age patients, proved to be more accurate than serum immunoglobulin for H. Pylori.*

KEYWORDS : Non-invasive tests, H. Pylori Infection, Upper GI Endoscopy

Introduction: -

Helicobacter Pylori (H. Pylori) infection is a worldwide problem and human being has been the preferred host colonized for at least 50,000 years and probably throughout their evolution. The organism colonizes from childhood and persists throughout life[1]. Extent of infection among developing nations is higher than in industrialized nation 80-90% Vs 25% respectively probably due to poor sanitary conditions and standard of hygiene[2].

Although most of the severe clinical manifestations of this infection appear in adults, the epidemiological studies showed that an acquisition of infection usually occurs in childhood. In paediatric age group, H. Pylori infection remains asymptomatic in most of the cases & in symptomatic cases it presents as gastritis. Symptomatology includes recurrent vomiting, recurrent abdominal pain, nausea, regurgitation, dyspepsia and hematemesis.

H. Pylori infection can be diagnosed by invasive and non-invasive tests[3]. The non-invasive tests include serologic tests, urea breath test and stool antigen assays. The invasive test comprises of endoscopy with biopsy analysis (histologic analysis and rapid urease test or culture of gastric antrum biopsy specimen) which is the gold standard test for diagnosis of H. Pylori infection[4-5]. Invasive tests are difficult to perform in paediatric population, so usually treating physicians prefer to do the non-invasive tests in children. The invasive test has a very high sensitivity (96%) and specificity (98.8%), even though it requires expertise for interpretation[6].

This prospective clinico-epidemiological study was conducted with an objective to compare the non-invasive test (Stool antigen for H. Pylori and Serum Immunoglobulin G for H. pylori) with invasive gold standard test (endoscopic biopsy and histopathology).

Method:-

The present study was a prospective cohort study carried out in the Department of Pediatrics, Umaid Hospital for Women & Children, Dr. S.N. Medical College, Jodhpur, over a period of twelve months.

The required sample size was 96 with a desired allowable error 10%, Power of study 80%, level of significance 0.05 and prevalence 60%. We enrolled 100 patients (54 males, 46 females) with gastro-intestinal symptoms (abdominal pain, nausea, vomiting, dyspepsia, hematemesis and diarrhoea) from outdoor and indoor wards of Department of Pediatrics, Dr S.N. Medical College, Jodhpur who fulfilled the inclusion criteria.

The study was approved by ethics committee of Dr S.N. Medical College. Upper gastrointestinal endoscopy (Olympus Video-scope (CV-15/GIF-XP 150 W) was performed in all enrolled cases after taking consent from parents. Biopsy sample was taken from gastric antrum and preserved in formalin and later sent to Department of Pathology, Dr S.N. Medical College, Jodhpur for histopathological examination (H&E Staining). Patients were considered H. Pylori positive when histopathology was suggestive for H. Pylori changes in antral mucosa.

We have also done Serum Immunoglobulin G for H. Pylori and Stool antigen for H. Pylori in all enrolled cases. We have used the SD BIOLINE H. Pylori Antigen Rapid Card test kit for Stool Antigen H. Pylori in which

test kit result window has 2 pre-coated lines, "T" (H. Pylori Antigen Test Line) and "C" (Control Line). Both the Test Line and Control Line in result window are not visible before applying any sample. The Control Line is used for procedural control and should always appear if the test is performed correctly. The SD BIOLINE H. Pylori Antigen Rapid Test kit can identify H. Pylori Antigen in human faecal specimen with high degree of sensitivity and specificity. It is a qualitative antigen detection test.

Serological IgG Test (Instant view H. Pylori rapid test) is a lateral flow immunoassay test for rapid, qualitative detection of IgG antibodies specific to H. Pylori in serum.

Sensitivity, specificity, positive predictive value and negative predictive value were calculated to evaluate the accuracy of non-invasive tests. Statistical analysis was performed with χ^2 test and student t test. The results were considered significant if p value is < 0.05.

Results: -

In our study, 23% cases (23/100) were positive for H. Pylori by histopathological examination of antral biopsy specimens (Table-1). Details of clinical presentation of enrolled cases are incorporated in table no. 1.

No significant differences were observed between H. pylori positive and negative cases in terms of demographic profile, anthropometry and clinical presentation (p>0.05).

In our study, among 100 cases 16 were positive for stool antigen H. Pylori and serum immunoglobulin G for H. Pylori, 6 cases were positive for stool antigen H. Pylori (Total-22) but negative for serum immunoglobulin G for H. Pylori, 3 cases were positive for serum immunoglobulin G H. Pylori (Total-19) but negative for stool antigen H. Pylori and 75 cases were negative for both stool antigen H. Pylori and serum immunoglobulin G H. Pylori.

On endoscopic examination, 43.47% H. Pylori positive cases had antral hyperaemia with or without gastro-oesophageal junction hyperaemia. 26.09% H. Pylori infected patients had antral nodularity with or without gastro-oesophageal junction hyperaemia as endoscopic finding. One H. Pylori infected case showed normal finding on endoscopic examination (Table-2).

Among 77 non-infected cases, 39 cases had normal endoscopic findings whereas 12 had gastro-oesophageal junction hyperaemia, 7 had villous atrophy and only 1 patient had antral nodularity.

On histopathological examination of antral biopsy specimen, 73.91% H. Pylori infected cases showed chronic H. Pylori gastritis (lymphoplasmacytic infiltration) whereas 26.09% cases had acute H. Pylori gastritis (neutrophilic infiltrations).

Stool antigen for H. Pylori had a sensitivity of 91.30%, a specificity of 98.70%, a positive predictive value of 95.45% and a negative predictive value of 97.43% while serum immunoglobulin G H. Pylori was 73.91% sensitive, 97.40% specific and had a positive predictive value of 89.47% and a negative predictive value of 92.59% and combination (in series) of non-invasive tests revealed a sensitivity of 69.56%, a specificity of 100%, a positive predictive value of 100% and a negative predictive value of 91.66% (Table-3).

In our study serum immunoglobulin G H. Pylori was positive in 73.91% cases who were H. Pylori infected whereas it was positive in 2.59% H. Pylori non-infected cases. The stool antigen was positive in 91.30% H. Pylori infected cases where as it was positive in 1.29% H. Pylori non-infected cases.

Difference in diagnostic accuracy between stool antigen H. Pylori and serum immunoglobulin G H. Pylori was statistically not significant (p>.05).

All patients (16/16) who were positive for both of the non-invasive tests, were found to be infected with H. Pylori on histopathological examination. Only one patient who was negative for both stool antigen H. Pylori and serum immunoglobulin G H. Pylori, showed positivity for H. Pylori on histopathology.

Association between H. Pylori positivity on histopathology of antral biopsy and combination of invasive tests (in series) was significant statistically (p <0.00001) i.e. if both stool antigen H. Pylori and serum immunoglobulin G H. Pylori showed positivity for H. Pylori, there are higher chances of H. Pylori positivity on histopathology of antral biopsy specimen (table-4).

Discussion: -

The prevalence of H. Pylori infection is high in developing countries across the world. Many invasive and non-invasive tests are available for detecting H. Pylori infection in current scenario. Upper gastrointestinal endoscopy and biopsy is considered as the gold standard test for diagnosing H. Pylori infection.

In the context of non-invasive tests, urea breath test is best available non-invasive test, but it has certain drawbacks such as high cost, limited availability and poor compliance in paediatric patients. In addition to these, its cut off value is not defined yet in paediatric patients.

The SD BIOLINE H. Pylori Antigen Rapid Card Test kit is a qualitative antigen detection test. It can identify H. Pylori antigen in human stool specimen with fair degree of sensitivity and specificity. The direct research of stool antigen is very simple, inexpensive, rapid and only one stool sample is required for testing. This also does not require an expert technician or expensive equipment and easy to perform. It could be used in epidemiological studies to determine the prevalence of H. Pylori infections in asymptomatic subjects.[7]

In general, H. Pylori prevalence is related to socio-economic status, hygiene and living conditions. A multicentre Italian study done to find out accuracy of stool antigen test for H. Pylori has revealed its utility in diagnosis and follow up of H. Pylori infection.[8,9] Like our study, many studies[9-13] done in past in paediatric patients have shown that stool antigen test had high sensitivity and specificity for diagnosing H. Pylori infection. The higher sensitivity, specificity, positive predictive value and negative predictive value of stool antigen for diagnosis of H. Pylori infection had shown that this test can be used for screening as well as diagnostic purposes.

Although the Instant View H. Pylori Rapid test, a lateral flow immunoassay test for rapid and qualitative detection of IgG antibodies specific to H. Pylori in serum, is quite specific for diagnosis of H. Pylori infection but it is less sensitive and has less PPV and NPV in comparison to testing for stool antigen for H. Pylori. Several workers also found serum IgG less useful in diagnosis of H. Pylori infection when compared to stool antigen.[9,13-16]

A positive co-relation of H. Pylori positivity on histopathology with positive results for both stool antigen and serum IgG H. Pylori was found in our study. Till now there is no literature available on comparison of combination (in series) of these two invasive tests with the endoscopy and histopathology. We found that there are more chances of positivity for H. Pylori on histopathology when both non-invasive tests showed positive results for H. Pylori and their association was significant statistically (p<0.00001).

Conclusion: -

Endoscopic biopsy and histopathology was the single most sensitive and specific test for diagnosis of H. Pylori infection. Stool antigen for H. Pylori had high sensitivity, specificity, positive predictive value and negative predictive value and it can be used both as a screening test as well as for diagnosis of H. Pylori infection in community.

Combination of these non-invasive tests had a diagnostic accuracy equivalent to gold standard test (endoscopic biopsy and histopathology). When facilities for upper gastrointestinal endoscopy are not available or the patients refused to go for endoscopy, then the combination of serology and stool antigen H. Pylori are sufficient enough to diagnose and treat a case of H. Pylori.

Table 1-: Clinical Presentation of Enrolled Patients

Clinical Presentation	Total (N = 100)	H. Pylori	
		Positive (n = 23)	Negative (n= 77)
Isolated Abdominal Pain	51	11 (21.57%)	40 (78.43%)
Abdominal Pain & Vomiting	17	05 (29.42%)	12 (70.58%)

Abdominal Pain & Diarrhoea	12	00 (0%)	12 (100%)
Abdominal Pain & Dyspepsia	10	05 (50%)	05 (50%)
Abdominal Pain & Hematemesis	02	01 (50%)	01 (50%)
Abdominal Pain, Vomiting & Dyspepsia	01	00 (0%)	01 (100%)
Isolated Vomiting	03	00 (0%)	03 (100%)
Vomiting & Dyspepsia	04	01 (25%)	03 (75%)

Table-: 2 Endoscopic Finding in H. Pylori Infected Patients

Endoscopic Finding	N = 23	%
Antral Hyperaemia	06	26.09%
Antal Nodularity	04	17.38%
Antral Hyperaemia with G. E. Junction hyperaemia	04	17.38%
Mucosal Erosion	03	13.05%
Mucosal Hyperaemia	03	13.05%
Antral Nodularity with G. E. Junction hyperaemia	02	08.70%
Normal	01	04.35%
Total	23	100%

Table 3-: Comparison of Sensitivity, Specificity, PPV, NPV of Individual Test & Combination of Non-Invasive Tests

Test	Parameters			
	Sensitivity	Specificity	NPV	PPV
Stool Antigen	91.30%	98.70%	97.43%	95.45%
Serum IgG	73.91%	97.40%	92.59%	89.47%
Serum IgG + Stool Antigen	69.56%	100%	91.66%	100%

NPV (Negative Predictive Value) PPV (Positive Predictive Value)

Table 4-: Diagnostic Accuracy of Non Invasive Test in Comparison to Histopathology

	Histopathology		Total (n=100)
	Positive	Negative	
Stool Ag Positive, S. IgG Positive	16	00	16
Stool Ag Positive, S. IgG Negative	05	01	06
Stool Ag Negative, S. IgG Positive	01	02	03
Stool Ag Negative, S. IgG Negative	01	74	75
Total	23	77	100

$\chi^2 = 85.959$ $p < 0.00001$

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