ORIGINAL ARTICLE

Histopathological Studies on Chronic Gastritis Associated with Helicobacter pylori Infection from Rural Area of India

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Abstract:
Background: Helicobacter pylori infection has been implicated as a major cause of chronic gastritis. It has been categorized as class 1 carcinogen. Aim and Objectives: An attempt has been made to evaluate the incidence of H. pylori in patients with chronic gastritis and to find out histopathologic indicators of presence of H. pylori in chronic gastritis. Material and Methods: A total of 101 biopsies from patients with chronic gastritis were evaluated using H & E and Giemsa stains. Biopsies were histopathologically graded according to the 1994 Updated Sydney system. Results: Neutrophilic activity was noted in 78 (77.2%), lymphoid aggregates were noted in 23 (22.7%) and atrophy was noted in 17 (16.83%) biopsies. The mononuclear cell infiltrate grading was mild in 43, moderate in 47 and marked in 11 biopsies. H. pylori were found positive in 79 (78.2%) biopsies. Neutrophilic activity with chronic gastritis and H. pylori association was highly significant (p value - 4.826x10⁻⁴). Significant association of presence of lymphoid aggregate with chronic gastritis and H. pylori (p value-0.0095) was also noted. Conclusion: Cases with chronic gastritis associated with neutrophilic activity with presence of lymphoid aggregate suggest H. pylori infection. Institution of rational treatment based on accurate diagnosis and eradication of H. pylori significantly reduces use of acid suppression and the risk for development of gastric malignancies.

Keywords: H. pylori, Chronic Gastritis, Histopathologic Examination

Introduction:
Helicobacter pylori are common cause of gastritis worldwide. Besides gastritis, H. pylori have been implicated to cause peptic ulcer, hyperplastic polyps, lymphocytic gastritis, anemia, peptic ulcer disease, duodenitis, intestinal metaplasia, gastric cancer and Mucosa-Associated Lymphoid Tissue (MALT) lymphoma. The diversity of clinical outcomes associated with H. pylori infection is probably a result of the interactions among host, environmental, and bacterial virulence factors [1].

Diagnosis of H. pylori can be done by both invasive and non invasive techniques. Non invasive techniques include serology, stool antigen test and carbon labelled urea breath test. The invasive techniques are rapid urease test, histology, polymerase chain reaction and H. pylori culture [2]. Special staining methods for detection of H. pylori can be used such as: Warthin-Starly; Cresyl-violet, Gimminez, Alcian yellow-toluidine blue, Genta stain, Giemsa stain and May Grünwald Giemsa [1].

The Sydney system updated in 1994 is the grading system of gastritis with which grading of gastritis can be done based on chronic inflammatory infiltrate, neutrophilic activity, lymphoid aggregate, metaplasia and presence of H. pylori infection using the visual analogue scale. In the present study, an attempt has been made to evaluate the incidence of H. pylori in patients with chronic gastritis and to find out histopathologic indicators of presence of H. pylori in chronic gastritis. To the best of our knowledge this is the first report from rural area of Marathwada region of India.
Material and Methods:
The present study is a cross-sectional study conducted over a period of 1 year. In all 101 cases presenting with symptoms of dyspepsia were included in the study which showed gastritis on histopathology. From the patients with dyspepsia underwent upper gastrointestinal endoscopy using flexible fibreoptic endoscope Fujinon – EG201FP, gastric mucosal biopsies were taken and immediately transferred to a container with 10% formalin. The biopsies were then sent to histopathology laboratory. After fixation, the specimens were subjected to tissue processing. The processed tissue was then embedded in paraffin to obtain 5µ thin sections. The sections were stained with Haematoxylin and Eosin stain and Giemsa stain for parasites [3]. In gastritis cases, the histopathological grading of gastric biopsies was done using the semiquantitative method of scoring according to the Updated Sydney system using the visual analogue scale. The histopathological variables density of *H. pylori*, neutrophil and mononuclear infiltration, atrophy and intestinal metaplasia were graded on a scale of 3 (mild, moderate and marked). Giemsa stain was used as a special stain for *H. pylori* detection. *H. pylori* colonization was assessed and graded after careful search for focal or complete involvement of the gastric surface. The sections were also examined for dysplasia or malignancy. For establishing association of different variables with incidence of *H. pylori* Chi square test and Fischer Exact test was applied.

Results:
A total of 101 biopsies with chronic gastritis were evaluated. Of these 60 biopsies were from antrum and 41 biopsies were from corpus. The mononuclear cell infiltrate grading was mild in 43 (42.57%), moderate in 47 (46.54%) and marked in 11 (10.90%) biopsies (Table1). *H. pylori* were positive in 79 (78.2%) biopsies (1+ grade of *H. pylori* in 36 (45.57%), 2+ in 25 (31.65%) and 3+ in 18 (22.80%) biopsies) (Fig.1) (Table 2).

<table>
<thead>
<tr>
<th>Particulars</th>
<th>Mild</th>
<th>Moderate</th>
<th>Marked</th>
</tr>
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<tbody>
<tr>
<td>Mononuclear cell infiltrate</td>
<td>43 (42.57%)</td>
<td>47 (46.54%)</td>
<td>11 (10.90%)</td>
</tr>
<tr>
<td>Neutrophilic activity</td>
<td>61 (78.2%)</td>
<td>15 (19.23%)</td>
<td>2 (2.56%)</td>
</tr>
<tr>
<td>Atrophy</td>
<td>15 (88.24%)</td>
<td>2 (11.76%)</td>
<td>00</td>
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</tbody>
</table>

<table>
<thead>
<tr>
<th>Particulars</th>
<th>Number of cases showing abnormalities</th>
<th>Number of cases showing <em>H. pylori</em></th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mononuclear cell infiltrate</td>
<td>101</td>
<td>79</td>
<td>78.2</td>
</tr>
<tr>
<td>Neutrophilic activity</td>
<td>78</td>
<td>73</td>
<td>93.58</td>
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<td>Lymphoid aggregate</td>
<td>23</td>
<td>23</td>
<td>100</td>
</tr>
<tr>
<td>Atrophy</td>
<td>17</td>
<td>14</td>
<td>82.35</td>
</tr>
</tbody>
</table>
Neutrophilic activity was present in 78 (77.2%) out of the total 101 biopsies of chronic gastritis. The grading of neutrophilic activity was mild in 61 (78.2%), moderate in 15 (19.23%) and severe neutrophilic activity was seen in only 2 (2.56%) biopsies with chronic gastritis (Table 1). In 73 (93.58) biopsies presence of both neutrophilic activity and \textit{H. pylori} was noted (Table 2). This association of presence of neutrophilic activity and \textit{H. pylori} was highly significant ($p$ value - $4.826 \times 10^{-4.826\times 10^{-4.826\times 10^{-4.826\times 10^{-4.826\times 10^{-4.826\times 10^{-}}}}}}$) (Fig. 2).

Lymphoid aggregate was seen in 23 (22.7%) biopsies of which four cases were associated with formation of lymphoid follicle. All the biopsies showing presence of lymphoid aggregate were associated with \textit{H. pylori} in 23 cases (100%) (Table 2) thus, highly significant association of presence of lymphoid aggregate with \textit{H. pylori} ($p$ value - 0.0095) was noted (Fig. 3).

Atrophy was noted in 17 (16.83%) biopsies of which 15 (88.24%) biopsies showed mild atrophy and 2 (11.76%) biopsies showed moderate degree of atrophy, marked atrophy was not seen in any of the chronic gastritis biopsies (Table 1). Atrophy and presence of \textit{H. pylori} was noted in 14 (82.35%) cases (Table 2) and the association was not significant using the Fischer exact test ($p$ value- 1)

Malignancy features were seen in three biopsies. Of which two biopsies showed diffuse type of adenocarcinoma and one biopsy showed signet ring cell type of carcinoma. All the three cases of malignancy were not associated with \textit{H. pylori} positivity.

\section*{Discussion}
\textit{H. pylori} contributes to gastro-duodenal injury by impairing local mucosal defense. This is caused by a very high urease activity which causes rapid hydrolysis of urea at intercellular junctions resulting in alteration in the milieu of the gastric epithelium preventing the normal passage of hydrogen ions from the gastric glands through the
mucosa to the lumen and instead permits back
diffusion. A consequence of back-diffusion is
hypochlorhydria and a predisposition to ulcer
formation [4].
The organism does not invade the tissue but lies
within the protective mucosal lining of the
stomach. Within the mucosal layer H. pylori
produces urease which assists in its survival. The
urease breaks down the urea present in gastric
juices into bicarbonate, carbon dioxide and
ammonia. Carbon dioxide and ammonia causes
belching and reflux. Ammonia neutralizes the
gastric acids allowing the organism to survive [5].
Individuals infected with H. pylori strains that
secrete Vac A are more likely to develop peptic
ulcers than people infected with strains that do not
secrete this toxin [1]. Infection with vacuolating
cytotoxin positive strains is reported to be
associated with particular gastroduodenal
diseases and was found to be more prevalent in
isolates of H. pylori from individuals with peptic
ulcer disease than those with gastritis. This offers
a potential explanation as to why only a small
percentage of persons infected with the organism,
develop gastro-duodenal ulcer [6]. Strains that
produce Cag A protein are associated with greater
risk of development of gastric carcinoma and
peptic ulcer [1].
Etiology of gastric cancer is multifactorial, most
commonly it develops after a long period of
atrophic gastritis [7]. Intestinal type of adeno-
carcinoma proceeds in a multistep manner and it
starts from the condition of chronic active
gastritis, followed by glandular atrophy, intestinal
metaplasia, dysplasia and finally gastric adeno-
carcinoma [8]. Three translocation are associated
with gastric MALToma, the t(11;18) (q21;q21),
t(1;14)(p22;q32) and t(14;18) (q32;q21) [9].
In the present study, grading of chronic gastritis
showed mild grade in 42.57%, moderate in
46.54% and marked in 10.90% biopsies. These
findings are slightly different from Mysorekar et
al. who showed mild grade of chronic
inflammation in 52.84%, moderate in 40.46% and
marked in 6.68% cases [10], however quite
similar to Suzana et al., who reported grade of
chronic inflammation was mild in 47.4%,
moderate in 40.90% and marked in 11.7% [1].
Neutrophilic activity of 77.2% is comparable to
73.38% of Suzana et al. [1].
Lymphoid aggregate of 22.7% in the present study
appears to be slightly higher than Mysorekar et
al. (30%) [10], quite higher than Suzana et al. (6.32%)
[1]. However, atrophy in 16.83% biopsies appears
to be quite similar to Suzana et al. (14.94%) [1],
quite higher than Mysorekar et al., who reported an
atrophy of just 1.67% [10]. In the present study, H.
pylori was seen in 78.2% biopsies, which fairly
correlates with Kumar et al. who reported a
prevalence rate 78% [11] and Mysorekar et al. who
reported prevalence rate of 74% [10].
In 93.6% biopsies presence of neutrophilic
activity was associated with H. pylori positivity.
This association of presence of neutrophilic
activity and H. pylori was highly significant (p
value - 4.826× 10⁻¹²). This finding is slightly
different from Kumar et al. who reported an
association of H. pylori with neutrophilic activity
in 83% cases [11].
In the present study all the biopsies showing
presence of lymphoid aggregate were associated
with the presence of H. pylori in 23 cases (100%),
thus highly significant association of presence of
lymphoid aggregate with H. pylori (p value -
0.0095) was noted. Mysorekar et al. showed
significant association of H. pylori with lymphoid
aggregate with germinal centre but the association
was not significant between H. pylori and
lymphoid aggregate without germinal centre [10].
Atrophy and presence of H. pylori was noted in 14
(82.35%) cases and the association was not significant using the Fischer exact test (p value - 1). Mysorekar et al. reported 5 cases of atrophy and all were associated with H. pylori but the association was not statistically significant [10]. Giemsa stain was used in this study as a special stain for H. pylori; this stain was found to be superior in H. pylori detection even when H. pylori were few in number. Also its cost effectiveness and ease of use makes it the best stain for H. pylori detection. It has been reported to be the best for H. pylori detection by Mysorekar et al. [10] and Kumar et al. also found it better than Warthin Starry stain and H & E stain [11].

Conclusion:
The results of present study indicate that Helicobacter pylori is strongly associated with chronic gastritis. Neutrophilic activity and presence of lymphoid aggregates in chronic gastritis cases was associated with H. pylori positivity, thus presence of these features can be taken as an indirect evidence of presence of H. pylori. It further indicates that finding of regional H. pylori prevalence and identifies high risk population infected with H. pylori helps to plan and implement treatment strategies in such patients to reduce complications associated with H. pylori infection.

References

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