

Prevalence of *Helicobacter Pylori* Gastritis at the North of Jordan

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Abstract

Helicobacter pylori was isolated from different gastric patients at the north of Jordan. Cultural and histological studies revealed a positive *H. pylori* infection in 78% of the collected samples. Clinical diagnosis showed that 21.6% of *H. pylori* patients were suffering from gastroduodenitis. Histological examination of collected mucosa showed that 67% of *H. pylori* positive patients were having acute and chronic gastritis, whereas 18.3% and 15% of them were suffering from intestinal metaplasia and atrophy, respectively. So, the highest specificity was 84% which was seen in histology results compared to microscopy. However, 58% of infected persons were males and the highest incidence of infection was found in the age 25-35 years old. Isolated *H. pylori* cells were found sensitive to tetracycline, amoxicillin and clarithromycin with an MIC of 0.15, 0.12 and 0.015 µg/ml, respectively. © 2011 Jordan Journal of Biological Sciences. All rights reserved

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1. Introduction

Helicobacter pylori is recognized as one of the most common chronic bacterial infections affecting humans worldwide (Rauws *et al.*, 1988; Petersen and Krogfelt, 2003).

Infection of *Helicobacter pylori* is highly associated with the upper gastrointestinal tract such as duodenal and gastric ulcers, gastric adenocarcinoma and non-Hodgkin's lymphomas of the stomach (Martin, 1997; Peek and Crabtree, 2006). Duodenal ulcer occurs among persons infected with *H. pylori* which might contribute to chronic atrophic gastritis development which is considered a risk factor for adenocarcinoma of the stomach (Martin, 1997). The role of *H. pylori* gastritis in ulcerogenesis and carcinogenesis was reported by Solcia *et al.* (1994). The most important virulence factors in *H. pylori* disease are believed to be: its motility, mucinase activity, urease production, adherence factors, heat-labile cytotoxins, hemolysin and lipopolysaccharide, in addition to its glycocalyx (Figura *et al.*, 1989; Geis *et al.*, 1989; Dunn *et al.*, 1990; Daw *et al.*, 1991; MacColm *et al.*, 1994; Patrick *et al.*, 1994; Petersen and Krogfelt, 2003).

Eradication of the pathogen can be achieved by triple regimen comprising bismuth, metronidazole and an antibiotic such as tetracycline or penicillin (Logan *et al.*, 1991). If metronidazole resistant strains are present, eradication of the pathogen can be achieved with omeprazole and amoxicillin or bismuth and ciprofloxacin. Monotherapy with clarithromycin was found effective (Logan *et al.*, 1991; Stenstrom *et al.*, 2008). This study reports the incidence of *H. pylori* gastritis at the north of Jordan.

2. Materials and Methods

2.1. Sample collection and preparation

Two biopsy specimens each were taken from sixty patients suffering from gastritis and referred for gastroscopy at the endoscopy unit at princes Basma hospital-north of Jordan. At least one of the biopsy specimens was taken from the corpus or the antrum or corpus and antrum of the patient's stomach. All biopsy specimens were taken from patients who had not been treated with bismuth compounds, antibiotics, H₂-receptor blockers or proton pump inhibitors but who showed gastrointestinal illness.

Specimens were collected in brucella broth containing 0.5% bovine serum albumin. They were transported in an ice box to the laboratory for immediate testing and culturing.

2.2. Organism and growth conditions

Biopsy specimens were removed from transporting medium using sterile forceps and 100 µl transport medium was added to the tissue. Then they were ground in a glass tissue grinder and inoculated into blood agar base supplemented with 7% human or horse blood, to which the following antibiotics were added: 10 mg/l vancomycin, 6mg/l amphotericin B and 5 mg/l trimethoprim (Sandra *et al.*, 1999). Mueller-Hinton agar was used to support the growth of *H. pylori* after the addition of 10% fetal calf serum. Incubation was done at 37°C under microaerophilic environment (BBL Campypack 71034) inside an anaerobic CO₂ jar for up to 7 days.

2.3. Identification of *H. pylori*

Morphological, cultural and biochemical characteristics of *H. pylori* were carried out according to Clidna and

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Julie, 1987; Natale *et al.*, 1989 and Leunk and Johnson, 1988.

2.4. Histological examination

All clinical specimens were processed for histopathological examinations using hematoxylin and eosin stain and Giemsa stain as described by Albertson *et al.*, 1998.

2.5. Statistical analysis

Results of diagnostic techniques were statistically compared using Chi-square analysis.

3. Results

Biopsy samples from sixty patients suffering from gastritis were collected. 62% of patients were males and 38% females, ranging from 23 to 94 years of age. All biopsy specimens were tested using microscopical, cultural and histological methods.

Out of the sixty patients, 47 gave positive cultures of *H. pylori* and the organism was isolated from both antral and corpus biopsies from 53 % of these positive patients (Table 1). Twenty four patients showed positive microscopical examination for *H. pylori*.

Clinical diagnosis showed that 21.6% of *H. pylori* patients were suffering from gastroduodenitis (Table 2). However, 15% of these patients developed gastric and duodenal ulceration while, 16.6% of *H. pylori* positive patients were diagnosed with atrophic gastritis (Table 2).

Histological examination of patient's mucosa showed three different abnormalities: Acute-chronic gastritis (neutrophilic and lymphocytic infiltration), intestinal metaplasia (replacement of gastric mucosa with intestinal mucosa) and gastric atrophy (thinning of gastric mucosa, loss of glandular tissues, and loss of parietal cells). As presented in Table3, 67% of *H. pylori* positive patients were having acute and chronic gastritis, whereas 18.3% and 15% of them were suffering from intestinal metaplasia and atrophy respectively.

Biopsy specimens showed polymorphnuclear and round cell infiltration (Fig1a). However, *H. pylori* was shown to colonize the gastric antrum cells (Fig1b).

The highest incidence of *H. pylori* among ages was those ranging from 25-35 years compared to other ages as

shown in Figure 2. Isolated *H. pylori* cells were found sensitive to tetracycline, amoxicillin and clarithromycin, when tested using an agar well diffusion method with an MIC of 0.15, 0.12 and 0.015 µg/ml, respectively.

4. Discussion

The prevalence of *Helicobacter pylori* differs significantly both between and within countries, with high rates of infection being associated with low socioeconomic status and high densities of living. (Goodman and Cockburn, 2001; Hazel and Francis, 2002). Approximately, 40 and 80% of adult individuals in developed and developing countries are infected respectively (Timothy and Martin, 1995). However, the percentage of infected people increases with age, since 50% of infected persons were those over the age of 60 compared with around 10% between 18 and 30 years (Pounder and Ng, 1995). But this was not the case in this study, since the highest percentage of patients was among young people ranging from 25-35 years old (Fig 2). In a large French cross-sectional study, a significantly lower prevalence of *H. pylori* infection was observed in females as compared with males (Broutet *et al.*, 2001). However, in this study a highest range of infection was found among males as shown in Figure 2.

In this study 78% of symptomatic patients were infected with *H. pylori*. The infection was associated with variable gastrointestinal illness, chronic gastritis, intestinal metaplasia and atrophic gastritis (Table 2). This is in agreement with others who reported that chronic superficial gastritis associated with *H. pylori* infection is a significant predisposing factor for the development of peptic ulcer, atrophic gastritis, gastric lymphoma and gastric adenocarcinoma (Martin, 1997; Alberto and Mario, 1998).

The highest specificity was 84% which was seen in histology results compared to microscopy (Table 1) which is comparable with Simor *et al.*,(1990) . Isolated *H. pylori* was found sensitive to clarithromycin, tetracycline and amoxicillin and their MICs were comparable to others findings (Pavicic and Namavar1993; Alistair, 1997). A follow-up incidence of *H. pylori* among different ages for the following years will be of importance.

Table 1. Statistical comparison between the three techniques used in the diagnosis of *Helicobacter pylori*.

Test kind	P _{value}	Sensitivity	Specificity	False positive	False negative
Microsc.-Culture	0.031	91.7%	30.6%	8.3%	69.4%
Histology-Culture	0.028	87.2%	53.8%	12.8%	46.2%
Histology-Microscopy.	0.031	46.8%	84.6%	53.2%	15.4%

Table 2. Clinical diagnosis of *H. pylori* positive patients after endoscopy.

Diagnosis	1*	2	3	4	2+3**	3+4	2+4
% <i>H. pylori</i> positive patients	10	15	21.6	16.6	25	6.8	5
(Number of patient)	(6)	(9)	(13)	(10)	(15)	(4)	(3)

*1 gastritis, 2 gastric and duodenal ulceration, 3 gastroduodenitis, 4 atrophic gastritis .

** .case repetition and percentage values to be considered.

Table 3. Histological results of *H. pylori* positive biopsy specimens.

	1- Acute and chronic gastritis	2-Intestinal metaplasia	3-Gastric atrophy
<i>H. pylori</i> positive patients	66.7%	18.3%	15%
(No. of patients)	(40)	(11)	(9)

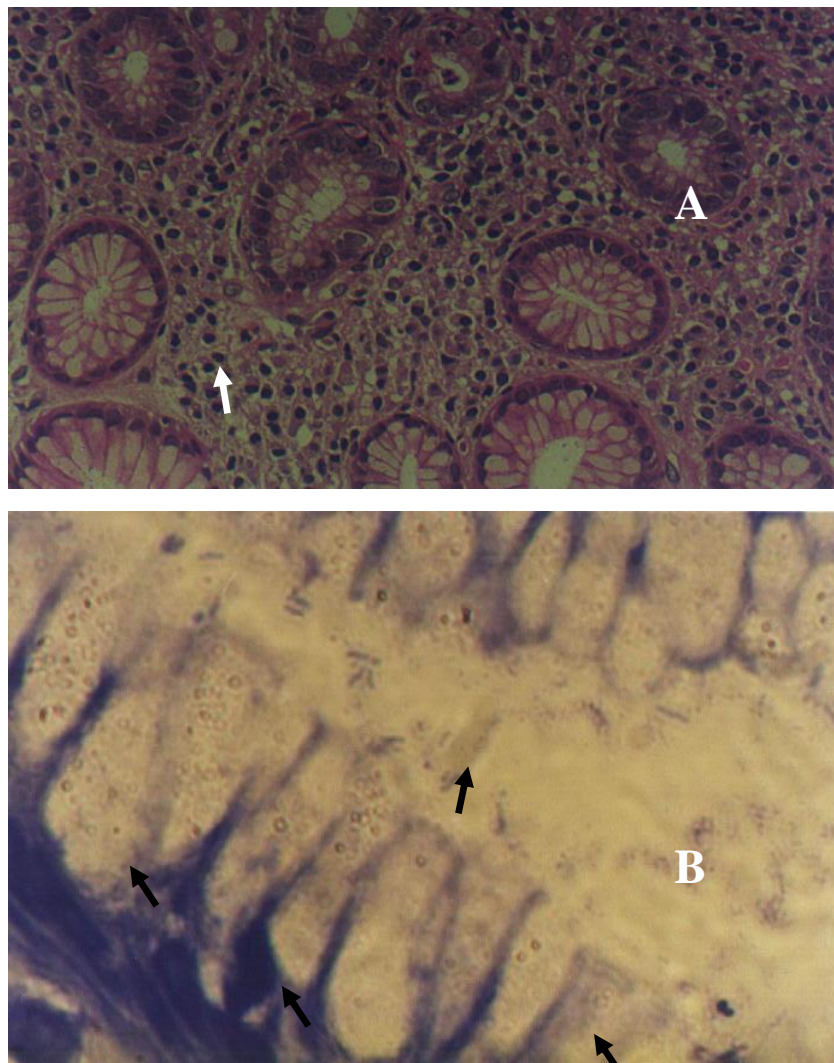


Figure 1. Photomicrograph of a patient gastric antrum infected with *H. pylori* stained with different stains . **A**, Hematoxylin and eosin stain, X 100 ; **B**, modified Giemsa ,X 100. Arrow indicates *H. pylori* cells.

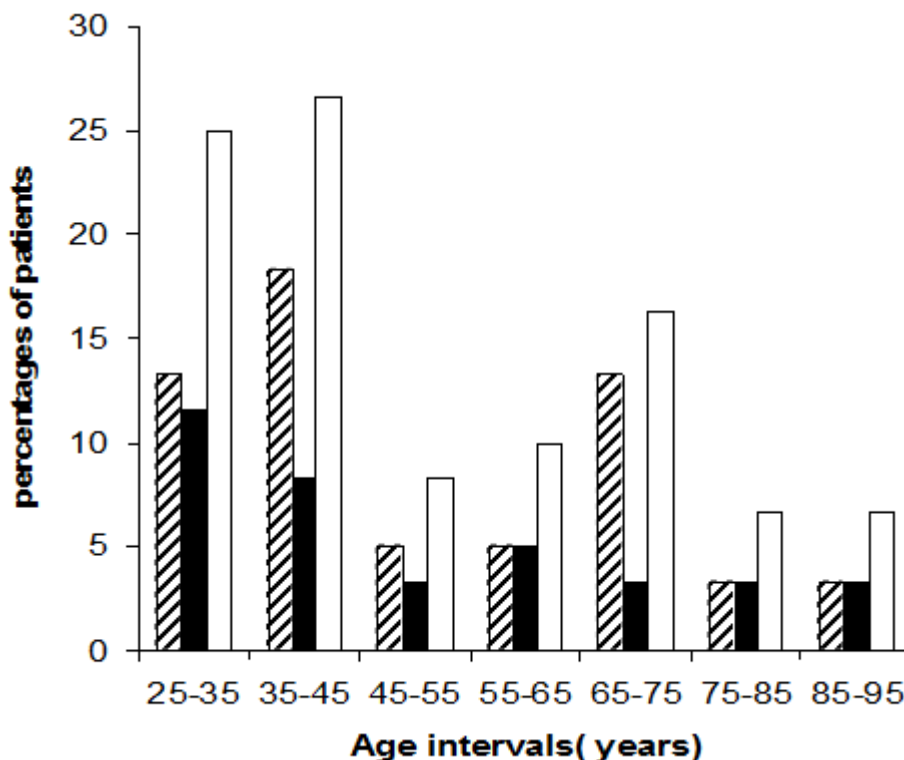


Figure 2. Distribution of *H. pylori* infection among different ages and sexes Male (▨) Female (■) Total (□).

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