

## PREVALENCE AND ANTIBIOTIC RESISTANCE AMONG *HELICOBACTER PYLORI* CLINICAL ISOLATES FROM MAIN HOSPITALS IN THE WESTERN REGION OF SAUDI ARABIA

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### ABSTRACT

**Objective:** The aim of this study was to evaluate the antimicrobial susceptibility patterns amongst *H. pylori* clinical strains isolated from the main hospitals in the western region of Saudi Arabia.

**Methodology:** Antimicrobial susceptibility testing was performed for 137 clinical isolates of *H. pylori* recovered from 368 Saudi patients undergoing endoscopic examination. The antibiotics used were amoxicillin, tetracycline, clarithromycin and metronidazole.

**Results:** A high percentage of resistance were observed against metronidazole (48.2%) followed by clarithromycin (27.7%), amoxicillin (14.6%) and tetracycline (9.5%). A total of 12 (8.8%) multidrug-resistant *H. pylori* isolates were observed in this study. Moreover, a warning sign of emerging resistance to amoxicillin, tetracycline and clarithromycin were noted.

**Conclusion:** The clinician need to be aware about resistance pattern in their region when they select empiric antibiotics regimen for *H. pylori*.

**KEYWORDS:** *Helicobacter pylori*, Antibiotic resistance, Gastritis, Peptic ulcer, Gastric cancer.

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### INTRODUCTION

*Helicobacter pylori* is recognized as a major cause of human gastritis and peptic ulcer as well as an important risk factor for gastric cancer. Eradication of this organism is

indicated in all patients with active or recurrent peptic ulceration.<sup>1</sup> Combined therapy (dual or triple) including two of the following antibiotics- amoxicillin, tetracycline, metronidazole or clarithromycin, plus a proton pump inhibitor (bismuth salt or ranitidine bismuth citrate) is the therapy most frequently used to eradicate *H. pylori*.<sup>2</sup>

During recent years, antibiotic resistance among *H. pylori* is considered the most common reason for eradication failure in most of the reports.<sup>2,3</sup> Several international studies demonstrated that high resistant rates among *H. pylori* clinical isolates especially metronidazole has emerged being a major factor for treatment failure.<sup>3,4</sup> However, resistance to some antibiotics such as amoxicillin is still absent or low.<sup>4</sup> Virtually, no treatment regimen has achieved a 100% cure rate, which makes it important to evaluate the antimicrobial susceptibility pattern of *H. pylori* strains in every geographical region.<sup>5</sup>

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The updated antimicrobial susceptibility pattern of *H. pylori* strains in Saudi Arabia and particularly in the western region is not available. Present study was therefore conducted to assess the prevalence of resistance among the four main antibiotics (amoxicillin, tetracycline, metronidazole and clarithromycin) currently in use for *H. pylori* infections treatment.

## MATERIALS AND METHODS

During October 2004 to May 2005, a total of 1104 gastric biopsies from 368 Saudi patients (3 biopsies from each patient) who presented with symptoms suggestive of chronic gastritis or peptic ulcer disease (PUD) were taken from antrum. Hospitals which participated in this study were; Al-Noor specialist hospital-Makkah (560 bed), King Abdul-Aziz hospital-Makkah (272 bed), General King Fahad hospital-Jeddah (710 bed), King Abdul-Aziz hospital and Oncology Centre-Jeddah (425 bed) and King Faisal hospital-Makkah (221 bed). Two hundred and eighty four (77.2%) of patients were suffering from various gastric pathologies, 264 (71.7%) with gastritis, 18 (4.9%) with gastric ulcer and two (0.5%) with tumor) while the remaining 84 (22.8%) had normal endoscopic results. Patients with a history of previous *H. pylori* treatment were excluded from the present study.

Gastric biopsies were transported in a 0.5 ml Brucella broth media (Oxoid, UK).<sup>6</sup> These gastric biopsies were obtained from each patient; one was used for rapid CLO test (Rapid Urease Test or *Campylobacter* like Organism for determination of urease activity) (Oxoid, UK) and the remaining two biopsies were cultured on *H. pylori* selective agar (Oxoid, UK) and incubated at 37°C in a BBL GasPak (Becton-Dickinson, USA) containing a Campy-Pak Plus microaerophilic system generator (Becton-Dickinson, USA) and incubated for 7 days. The identity of *H. pylori* clinical isolates were confirmed by colonial morphology, Gram-stain (curved Gram-negative bacilli) and positive reaction for oxidase, catalase & urease tests.<sup>6-8</sup>

Antibiotic susceptibility was examined by the disk diffusion method, according to a standard

protocol.<sup>9</sup> Briefly, bacterial suspensions were adjusted to the 0.5McFarl and standard (equivalent to  $1-2 \times 10^8$ cfu/ml) and were used to inoculate Muller Hinton agar plates. Antimicrobial disks (amoxicillin, tetracycline, metronidazole and clarithromycin) were applied and the plates were incubated under microaerophilic conditions at 35°C for 16-18 hour. The zones of growth inhibition produced by each antibiotic were measured and interpreted by standard procedure, to determine the susceptibility or resistance.

## RESULTS

This study included three hundred sixty eight patients i.e. 64 (46.7%) male and 73 (53.3%) female. Age ranged between 16-90 years, majority (34.3%) were between 31-45 years (Fig-1). With culture technique 137 (37.2%) were positive. In comparison to culture test the direct CLO test showed lower positive results with only 128 (34.7%) samples being positive. (Fig-1)

According to the endoscopic findings, the presence of *H. pylori* in normal, gastritis, gastric ulcer and malignant cases were 22.6% (31 out of 137), 70.8% (97), 6.6% (9), and 0%, respectively (Figure-2).

The antimicrobial resistance patterns of *H. pylori* isolates are shown in Table-I. Results demonstrated that among the tested isolates, 13 (9.5%), 20 (14.6%), 38 (27.7%), and 66

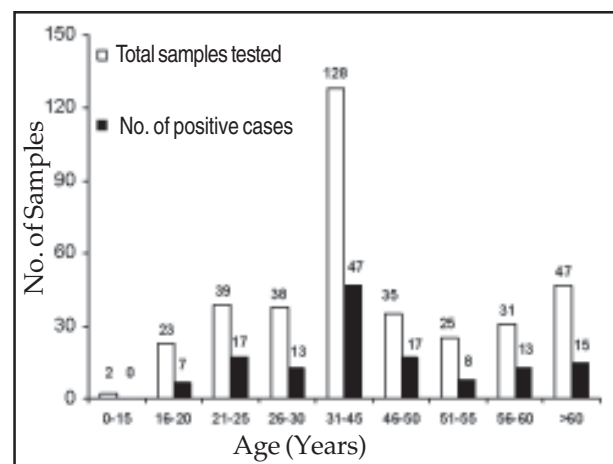


Figure-1: Distribution of *H. pylori* cases according to the age groups of the patients.

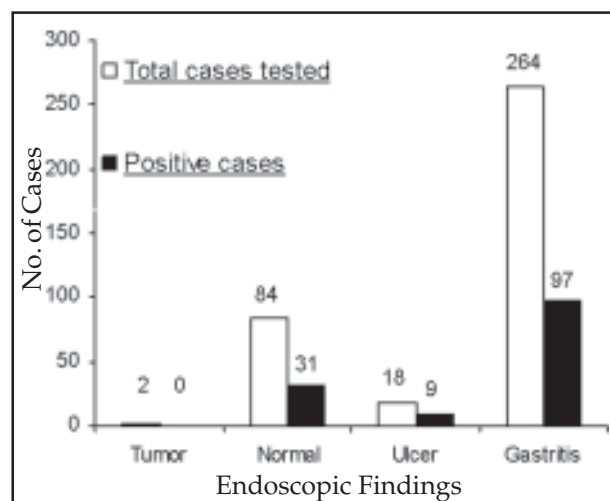


Figure-2: Distribution of positive cases in relation to endoscopic findings.

(48.2%) strains were resistant to tetracycline, amoxicillin, clarithromycin and metronidazole, respectively. Results revealed that a total of 12 (8.8%) *H. pylori* isolates were multidrug-resistant (i.e. resistant to metronidazole, clarithromycin and amoxicillin).

## DISCUSSION

Present study demonstrated 37.2% prevalence rate for *H. pylori* in the patient population tested. The results are much lower than compared to what has been reported elsewhere in Saudi Arabia with prevalence rates of 54.9% and 63% in southern and eastern regions, respectively.<sup>10,11</sup> This difference in prevalence rates could be attributable to the different detection methods in each study, different demographic distribution of the bacteria among various regions and previous antibiotic consumption.<sup>2,12-14</sup>

In this study, the prevalence rate of *H. pylori* in patients with gastritis was 70.8% which is much higher when compared to the results

obtained by Ayoola *et al.*, reporting a percentage of 55%.<sup>10</sup> The prevalence of *H. pylori* among gastric ulcer and control cases in this study were 50% and 22.6%, respectively, which is comparable with the results achieved by Ayoola *et al.*, who reported a percentage of 53.6% and 43.6% in gastric ulcer and normal cases, respectively.<sup>10</sup> Interestingly, two earlier studies from Saudi Arabia detected the organism in only 9% and 13%, respectively, in normal controls in comparison to 22.6% obtained in the present study, reflecting an increase in the carrier rate of this organism.<sup>15,16</sup>

The status of metronidazole resistance (48.2%) obtained in this study is much lower than what has been reported by Al-Qurashi *et al.* in other region of Saudi Arabia who reported an increase of metronidazole resistance among *H. pylori* clinical isolates from 35.2% to 78.5% in period between 1988 to 1996.<sup>17</sup> However the rate of metronidazole resistance in our study is much higher than what has been reported in other studies with resistance percentage of 16% to 90%.<sup>2,3,18-21</sup> The variation in the resistance rates obtained in this study compared to other studies may indicate the differences in metronidazole misuse in different regions. Resistance to clarithromycin was significantly higher in this study (27.7%) compared to the recent reports from the United States (13%) and Italy (23.4%).<sup>19,20</sup>

This study showed that 13 isolates were resistant to tetracycline in comparison to only a single isolate in other region of Saudi Arabia in a study performed in the period 1990-1996 and none in the period 1987-1988 indicating a gradual tetracycline emerging resistance among *H. pylori* clinical isolates.<sup>17</sup> Similarly, the resistance rate to amoxicillin in this study (14.6%) was much higher than what has been

Table-I: Distribution of antibiotics resistance of *H. pylori* according to the endoscopic findings.

Endoscopic Findings	Rate of Resistance (%)			
	Amoxicillin	Clarithromycin	Tetracycline	Metronidazole
Gastritis (n=97)	11 (11.3)	24 (24.7)	8 (8.2)	40 (41.2)
Ulcer (n=9)	2 (22.2)	2 (22.2)	1 (11.1)	7 (77.8)
Normal (n=31)	7 (22.6)	12 (38.7)	4 (12.9)	19 (61.3)
Total (137)	20 (14.6)	38 (27.7)	13 (9.5)	66 (48.2)

reported elsewhere.<sup>12,18,22</sup> We conclude that, the clinicians need to be aware about antibiotic resistance pattern in their region when they select empiric antibiotics regimen for *H. pylori*.

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