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Serological and Epidemiological Studies of *Helicobacter pylori* Infection at District Mardan Based on Gender and Different Age Groups

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Abstract

The current study was aimed to observe the prevalence of *Helicobacter pylori* infection at District Mardan with respect to their gender and age groups. The blood samples were collected from all the subjects who attended the OPD at Mardan Medical Complex. All these samples were screened for the presence of antibodies against *H. pylori* through a rapid antibody assay. Positive and negative cases were recorded based on the test results and prevalence was measured keeping in mind the selected parameters in individuals of each group. A total of 4312 people was included in the study, out of which 2586 were males and 1726 were females. Majority of the patients were in the mature age group. Based on our results, there were 125 males demonstrating a prevalence of 4.83% which is slightly higher than 4.66% prevalence of 77 females infected. Comparisons were made based on gender and age groups and data presented in the form of figures and tables. Monthly wise prevalence has also been shown. Moreover, prevalence was higher in the people aged 19-44 and lowest in immature ones. However, the overall prevalence in the subjects was 4.68% for the span of 4 months.

Keywords: *Helicobacter pylori*; Seroepidemiology; Prevalence; Pakistan

Introduction

Helicobacter pylori are spiral shaped gram-negative bacteria which infects more than 30% population worldwide and, in few countries, this infects more than 50% people of the total population. Marshall and Warren were awarded Nobel Prize in 1982 for the discovery of this bacterium. It is one of the most studied bacteria and over 29000 articles established its importance in human disease. This bacterium is responsible for gastritis, gastric and duodenal ulcer and gastric cancer [1]. Globally different strains of *H. pylori* seem to be associated with differences in virulence, and the resulting interplay with host and environmental factors leads to subsequent differences in the expression of disease. Age, ethnicity, sex, geography and socioeconomic status are all factors that influence the incidence and prevalence of *H. pylori* infection. In developing countries, *H. pylori* infection is a public health

issue. The high prevalence of the infection means that public health interventions may be required. Therapeutic vaccination is probably the only strategy that would make a decisive difference in the prevalence and incidence of *H. pylori* throughout the world. The overall prevalence is higher in developing countries and lower in developed countries and within areas of different countries. There may be similarly wide variations in the prevalence between more affluent urban and rural populations. The principal reasons for these variations involve socioeconomic differences between populations. Transmission of *H. pylori* is largely by the oral-oral or faecal-oral routes. Lack of proper sanitation, safe drinking water, basic hygiene, poor diets and overcrowding all play a role in determining the overall prevalence of *Helicobacter pylori* infection [2]. In 1994, *Helicobacter pylori* was categorized as a class I human carcinogen by the WHO International Agency for Research on Cancer (IARC) due to its epidemiological connection to gastric cancer [3].

In Pakistan, acid peptic disease due to the prevalence of *Helicobacter pylori* infection is very high among population and the number of patients is continuously increasing mainly due to non-availability of ideal diagnostic and treatment facilities in public healthcare sector. As *H. pylori* is a rising issue in Pakistan, so according to experts of health *H. pylori* infection may become chronic with the chances of gastric carcinoma and stomach cancer, if it is not treated fully and properly [4].

The existence of spiral shaped micro-organisms like *H. pylori* in the stomach of human was observed at least 100 years before by Prof W. Jaworski who was a Polish Clinical Researcher at the Krakow Jagiellonian University. Health experts believe that in order to decrease the chances of stomach cancer, *Helicobacter pylori* infection should be treated properly and the bacterium should be eliminated completely from the body. Half population of the world is affected with *Helicobacter pylori* infection but still the way of transmission has not been completely clarified. There are some indications in order to propose that it is transmitted from individual to individual through the fecal oral way but still the way of transmission is undecided and faecal to oral or oral to oral are the most commonly routes of transmission at this phase. Although most infections happen in childhood, *Helicobacter pylori* infect both male and female equally. The presence of *Helicobacter pylori* in stomach persuades a chronic, active, inflammation in approximately every individual who is infected. Majority of the individuals with *Helicobacter pylori* are, however, asymptomatic and there are less than 10% of persons with *Helicobacter pylori* in which appear peptic ulcer disease, Mucosa Associated-Lymph-Tissue (MALT) lymphoma or gastric cancer. Symptoms of H. pylori in anybody appear when it damages his stomach's lining or top part of his duodenum [5].

Materials and Methods

Study site

This present study was conducted from 1st of September 2017 to 30th December 2017 at Mardan Medical Complex and Teaching Hospital, KP with an aim to assess the prevalence of *Helicobacter pylori* infection across population in district Mardan.

Data and samples collection

A total of 4312 patients which included referrals from both outpatient and hospital inpatient services were taken onto consideration. Among them, 2586 were males and 1726 were females. All the patients were further sub-divided into three groups based on their age, i.e. immature, mature and adults. Blood samples were taken and sent to pathology laboratory for further analysis. These patients were included only once in the process.

H. pylori test

Blood antibody test named as *H. pylori* Ab rapid assay was carried out to assess antibodies formed against *H. pylori* bacteria in the serum. Approximately 30 µl drop of serum was transferred into the test strips and one drop of 40 µl buffer was added. After few minutes, a coloured line appeared and results were recorded. Two distinct red lines appear, one line in the control region (C) and second in test region (T), showed positive results. Appearance of red line only in the control region (C) and not in the test region demonstrated negative results. The concentration of red colour in the test line region (T) varies depending on the conception of *H. pylori* antibodies in the sample. Hence, any shade of red colour in the test region (T) was reflected as positive.

Data analysis

Statistical analysis of the data was performed using MS Excel. Percentages and values were presented in the form of figures and tables and comparisons were made among them.

Results

All the data has been presented in the form of figures and tables to illustrate the number of normal and infected individuals, prevalence on monthly and gender basis and also comparison in different age groups. In this study, a total of 4312 patients were screened for anti *H. pylori* (*Helicobacter pylori*) antibody by rapid assay (serum method). It was found that 202 patients were positive for *Helicobacter pylori* infection as depicted in **Figure 1** and **Table 1**, with an overall prevalence of 4.68%. **Table 1** illustrates that there are 125 male patients positive for anti *H. pylori* antibody with the prevalence value of 4.83% and among them, 11 are immature patients having age of 3-18 year are *Helicobacter pylori* positive which show 4.18% prevalence in immature male

patient, while 78 mature male patients of the age 19-44 years are positive for *Helicobacter pylori* infection which show 5.49% and 36 male adults having the age of 45-65 are positive for *Helicobacter pylori* infection which show 3.98% prevalence.

Table 1: Number and *Helicobacter pylori* prevalence of normal and infected people of different genders and varied ages.

	Type	Age (in years)	Subjects	Normal	Helicobacter pylori +	Prevalence (%)
	Immature	03-18	487	470	17	3.5
	Mature	19-44	2402	2274	128	5.32
Overall	Adult	45-65	1423	1366	57	4.05
Total			**4312**	**4110**	**202**	**4.68**
	Immature	03-18	263	252	11	4.18
	Mature	19-44	1420	1342	78	5.49
Males	Adult	45-65	903	867	36	3.98
Total			**2586**	**2461**	**125**	**4.83**
	Immature	03-18	224	218	6	2.67
	Mature	19 - 44	982	932	50	5.09
Females	Adult	45-65	520	499	21	4.03
Total			**1726**	**1649**	**77**	**4.66**

Out of 202 positive cases, 77 are female positive patients in a total of 1726 females as obvious from **Table 1**, which show 4.66% prevalence. Among them, there are 6 immature female patients in the age of 3-18 which is 2.67%, while out of 77 female patient just 50 number of patient are positive in the mature female in the age of 19-44 which show 5.09% prevalence, and just 21 are positive in the adult female their age is 45-65 which show 4.03% prevalence.

From **Figure 1**, it is clear that in the month of January, total patients were 1208 and out of them 59 were infected with *Helicobacter pylori* showing the prevalence of 4.88%, while in February, 812 patients are registered and out of them, 21 were positive having prevalence are 2.58%.

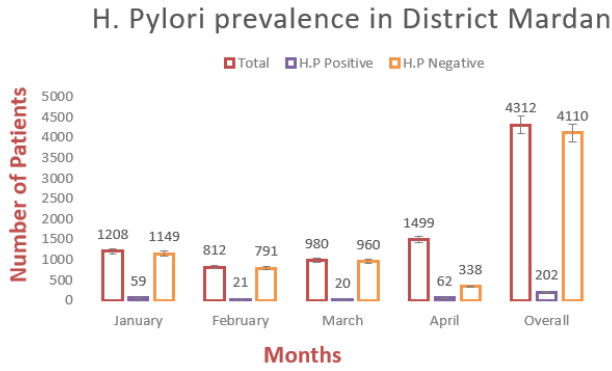

Figure 1: Number of *Helicobacter pylori* infected individuals among patients in Mardan during the span of 4 months.

In the following month of March, total number of patients are 980 and number of cases confirmed for *Helicobacter pylori* were 20 and the prevalence ratio are 2.04%. In April, total positive patients were 62 out of 1499, which shows 4.14% prevalence. All this information is available both from **Figures 1 and 2**.

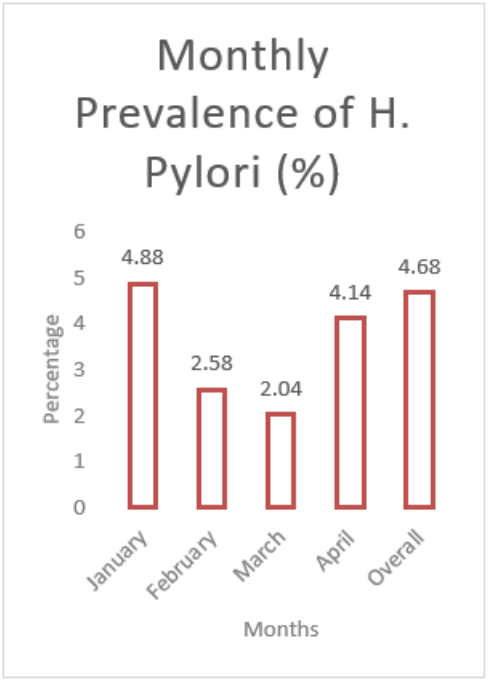

Figure 2: Percent prevalence on monthly basis.

Moreover, **Figure 3** depicts the prevalence and percentage of infected and normal individuals in percent based on different age groups regardless of the gender. *Helicobacter pylori* prevalence is higher i.e. 5.32 in mature individuals followed by adults and then immature individuals.

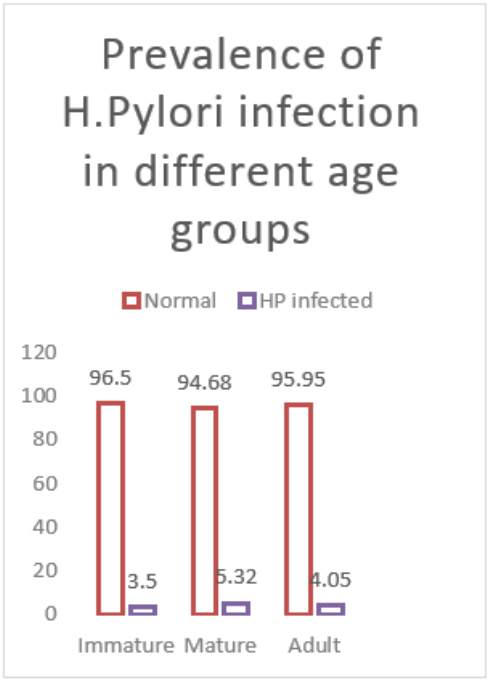

Figure 3: Percent of normal and infected individuals in different age groups.

Discussion

Helicobacter pylori is a harmful gram-negative bacterium infecting more than 30% population of the world and in few countries, above 50% of people are victims of the disease due to *H. pylori* infection [6]. *H. pylori* is considered to be the leading cause of human gastritis, duodenal and gastric ulcers and has been categorized as class-1 human carcinogen [7]. Usually the disease progress through gastritis as a result of *H. pylori* infections followed by atrophy, intestinal metaplasia and dysplasia that can cause gastric mucosal carcinoma. Gastritis is developed in almost 90% individuals infected with *H. pylori* whereas gastric atrophy and intestinal metaplasia appear more often in *H. pylori* positive than in negative patients [8]. Genetic diversity and origin of the infecting *H. pylori* strain are the major factors that determine the outcome of *H. pylori* disease in most cases [9]. Virulent *H. pylori* cytotoxic-associated gene A (cag A) positive strains are associated with more severe state as patients infected with cag A positive strains had higher risk of gastric cancer development than those of cag A negative strains. Various studies reported that 60%-70% of *H. pylori* strains isolated from North American and European populations had cag A gene [10]. A study conducted at Abakaliki by Ugwu had shown that majority of *Helicobacter pylori* infected patients were more than 60 years of age [11]. The percentage of active *H. pylori* infection in the Northern region of Pakistan showed 53% of prevalence [12]. A high rate of active *H. pylori* infection (74.4%) in asymptomatic subjects in a rural area of Islamabad Pakistan has been reported [9]. Several epidemiological reports have shown that the rate of *H. pylori* infection increases significantly with age, with more than 80% of children being infected by the age of 10 years [13]. The variation in the prevalence of *H. pylori* infection between different populations suggests that different parameters such as socioeconomic status and environmental factors play a key role in the positivity of *H. pylori* infection [14].

In the current study we have examined 4312 hospital patient and general OPD patient from January 2017 to April 2017 for Anti *H. pylori* antibodies patients who were coming from different regions of Mardan KP, Pakistan to Mardan Medical Complex. The patients were screened for Anti *H. pylori* infection by Ab rapid assay (serum method) which showed an overall prevalence rate of 4.68%. Out of total patients screened, 125 were positive in males and 77 in females for anti *H. pylori* antibody with the prevalence of 4. 83% and 4.465% respectively. In Mardan, most of the patient screening for detecting *H. pylori* is done by local laboratory which show inaccurate results. This large variation in percent prevalence of the present study compared to other studies at different regions of Pakistan could be due to the type of test type used to diagnose the disease, false negative cases and low standards of the pathology centre. Moreover, it is also likely that most of the patients go for check-up, diagnosis and treatment to private medical centres and labs and therefore are not registered in the hospital where we performed our study. So, we recommended that people must be use standard laboratory for the diagnosis of *H. pylori* infection and demand from government that health facility must be provided. Improvement in the treatment and diagnosis of *H. pylori* infection should be made. Also, aware the people of Mardan KP and all Pakistan about *H. pylori* infection and their precautionary measures. Patients of *H. pylori* infection records and serum of all patients were reported to and analyzed at the Mardan Medical Complex, Pakistan from September 2019 to December 2019.

Conclusion

The current study highlights the total number of patients screened in the given span of time to assess the prevalence of *Helicobacter pylori* infected individuals across Mardan region. No significant difference has been noticed based on gender in prevalence of *Helicobacter pylori*, although it varies to some extent in different aged individuals. There is need to further explore the transmission routes and risk factors of the disease in the area under consideration. Standard diagnostic and quality treatment in addition to prophylactic and preventive measures can prove as quality tools to eradicate this infection completely from the population.

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Authors Contributions and Consent

Haq I contributed to the main idea, conceptualization and performing the research work. Faisal S contributed in the data collection. Abdullah have contribution in formatting the manuscript draft and creating figures and tables. Asghar M, Afridi GZ, Zahir F, Iqbal S, Farkhanda, Khan A, Rehman AU, Ullah R, Ali F, Ullah H, Waqas M and Khan A have contribution in the statistical and critical analysis of the manuscript and also help in proofreading. All authors are agreeing to the publication of this article in Journal of Biomedical Sciences.

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Conflict of Interest

The authors declare no conflict of interest.

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