Seroprevalence of *Helicobacter pylori* Among Asymptomatic Individuals of Northern Pakistan: A Cross Sectional Study

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ABSTRACT

Helicobacter pylori is a causative agent of several gastric diseases and its presence in asymptomatic individuals is a continuous threat to acquire symptomatic diseases. The objective of the study was to investigate the seroprevalence rate and linked factors in asymptomatic individuals of northern districts of Pakistan. A cross-sectional study was conducted between November 2020 to August 2021 including 600 participants. Seropositivity was tested through *H. pylori* strip from collected serum and data was collected through questionnaire. Univariate analysis was carried out for association of factors with seropositivity, and associated factors were predicted using regression model. The *H. pylori* infection rate is highest in District Swat with 50% seropositivity found in asymptomatic individuals (p=0.002, OR: 0.398 CI 0.213 – 0.714). Out of the total tested individuals, 28.7% were found to be *H. pylori* positive. A significantly higher infection rate was observed in positive family members (80.0%, $p = 0.04$). A regression model gives an OR of 1.147 (CI: 0.942 - 1.396) of seropositivity in household members. A positive family member could be a potential source of transmitting *H. pylori* infection in individuals as the course of transmission is oral or oral-fecal. Therefore, studies on large cohort for investigation of seropositivity in household members would be beneficial.

Keywords: *H. pylori*, Seroprevalence, Asymptomatic, Northern Pakistan

1. INTRODUCTION

Helicobacter pylori, a gram-negative microaerophilic bacterium, infects almost 50% of the global population [1]. It colonizes the mucosal surface of the stomach and instigate inflammatory response causing diseases from mild gastritis to deadly gastric carcinoma and MALT (mucosa-associated lymphoid tissue) lymphoma [2,3]. Globally, there are millions of deaths annually owing to peptic ulcer and gastric cancer

[4,5] as the risk of developing carcinomas by this bug is three to six-fold [6,7]. The infection is transmitted by intra-familial oral-oral or oral-fecal routes [8] and most of the diseases come by during childhood with lifelong endurance [9].

Different nations have diverse rates of H. pylori prevalence, with Africa having the highest infection rate (79.1%). Regional prevalence studies showed 63.4% prevalence in Latin American and Caribbean people, 54.7% in Asian, 37.1% in Northern American, and 24.4% in Oceanian populations [10]. Despite a decline in infection rates in developed countries, its frequency is still high in underdeveloped countries, reaching up to 80% [11]. Age, gender, and ethnic group disparities in incidence have been observed in addition to geographic variances [12]. In Saudi Arabia, an epidemiological study reveals a high frequency of 35–67% in adults. An epidemiological analysis of gastric cancer patients over 10 years found a range of 0.03-4.91% in females and 0.04-14.87% in males. According to some other studies, the risk rises with age and occurs at a higher rate in women than in males. [13–15].

Overcrowding, low socioeconomic status, poor personal and environmental hygiene, poor quality of drinking water and food, are the principal risk factors for *H. pylori* infection transmission [16,17]. Despite a very high *H. pylori* infection prevalence in Pakistan i.e., 50-90%, a limited concern has been taken on *H. pylori* prevalence in non-symptomatic individuals and hence only a limited fundamental data on *H. pylori* prevalence is available. Assessing the regional prevalence of infection in asymptomatic individuals based on disease related features would somehow help in sorting of socio-demographically prone individuals. Consequently, this would facilitate disease detection at latency and its possible chances of onset based on immunological and socio-economic features. The current study is designed to assess the local prevalence of *H. pylori* infection in asymptomatic individuals as well as identification of associated factors from Northern areas of Khyber Pakhtunkhwa, Pakistan.

2. METHODOLOGY

2.1 Study Design and Participants

A cross-sectional study was carried out between November 2020 to August 2021 permitted by the Institutional Review Committee of Hayatabad Medical Complex (370/HEC/B&PSC/2020). A total of 600 individuals belonging to different localities and classes were included in the study.

2.2 Inclusion and Exclusion Criteria

Only those individuals were included in the study who did not have gastric complications at the time of data collection and had no history of gastric diseases. All those persons who had a history of any gastric

disease like gastric surgery, peptic ulcer, abdominal pain, dyspepsia as well as those taking non-steroid anti-inflammatory drugs were excluded from the study.

2.3 Sample Collection and Informed Consent

The written informed consent was collected from the studied participants. The information was collected directly by interviewing the participants and data was recorded on a structured questionnaire.

2.4 Serological Screening

Blood was taken in serum tubes from each participant and serum was separated. Commercial antibody strip (CITI Scientific supply, Karachi, Pakistan) was used to test *H. pylori* specific IgG antibodies from the sera. It was done by adding a drop of the serum sample and buffer on the absorbent pad of the strip. Appearance of clear test and control line indicates positive results while appearance on control line only indicates absence of *H. pylori.*

2.5 Data Analysis

The recorded data was organized in excel sheets and analyzed using IBM SPSS software (version 22.0). The frequency and percentage of demographic factors were evaluated, and chi-square was used for finding the association of those features with non-symptomatic positive individuals. Finally, univariate, and multivariate models were used to sort out the independent factors that can be used as predictors of infection.

3. RESULTS

The current observational cross-sectional study was directed with the aim to estimate prevalence and characteristics associated with *H. pylori* infection in non-symptomatic individuals. There were 29.2% (n = 175) positive individuals among 600 included participants. Most of the positive individuals were male (60.6%) and more than 50 years of age (28.0%) and 78.9% were married. No association of gender, age, and marital status was found with *H. pylori* positivity. The majority of participants in the cohort were uneducated (35.3%) and 57.8% live in a family with family members ranging from 6-10. Exploring educational and family status of the *H. pylori* positive individuals designate high frequency in the same groups i.e., uneducated (39.4%) and 6-10 family members (63.4%) as appeared in the cohort. There was no association of the investigated features with the infection state. District-wise distribution of subjects revealed an association of the district with seropositivity (p = 0.01) (Table 1).

Table 1: Characteristics of Subjects

Sociodemographic Factors		Descriptive Statistics			Univariate Test
Factors	**Categories**	**Total N (%)**	**Negative N (%)**	**Positive N (%)**	χ^2 **p-value**
Total		600	425 (70.8)	175 (29.2)	
Gender	Female	246 (41.0)	177 (41.6)	69 (39.4)	0.62
	Male	354 (59.0)	248 (58.4)	106 (60.6)	
Age group	11-20	30 (5.0)	21 (4.9)	9 (5.1)	0.41
	21-30	103 (17.2)	65 (15.3)	38 (21.7)	
	31-40	154 (25.7)	114 (26.8)	40 (22.9)	
	41-50	143 (23.8)	104 (24.5)	39 (22.3)	
	>50	170 (28.3)	121 (28.5)	49 (28.0)	
Marital Status	Married	490 (81.7)	352 (82.8)	138 (78.9)	0.25
	Single	110 (18.3)	73 (17.2)	37 (21.1)	
District	Buner	66 (11.0)	56 (13.2)	10 (5.7)	0.01
	Dir	108 (18.0)	96 (22.6)	12 (6.9)	
	Kohistan	49 (8.2)	43 (10.1)	6 (3.4)	
	Malakand	65 (10.8)	56 (13.2)	9 (5.1)	
	Shangla	111 (18.5)	61 (14.4)	50 (28.6)	
	Swat	201 (33.5)	113 (26.6)	88 (50.3)	
Family members	1-5	75 (12.5)	58 (13.6)	17 (9.7)	0.21
	6-10	347 (57.8)	236 (55.5)	111 (63.4)	
	11-15	114 (19.0)	81 (19.1)	33 (18.9)	
	>15	64 (10.7)	50 (11.8)	14 (8.0)	
Education	above matric	71 (11.8)	45 (10.6)	26 (14.9)	0.15
	below matric	109 (18.2)	84 (19.8)	25 (14.3)	
	Matric	128 (21.3)	97 (22.8)	31 (17.7)	
	Nil	80 (13.3)	56 (13.2)	24 (13.7)	
	Uneducated	212 (35.3)	143 (33.6)	69 (39.4)	

In addition to demographic features, the characteristics that can lead to disease progression like smoking, *H. pylori* infected patients in family, source of water, toilet use, and awareness were also investigated as tabulated in table (Table 2). Among all the above-mentioned attributes, the only factor that showed association with *H. pylori* positivity is their positive family members (80.0%, $p = 0.04$).

Table 2: Infection Related Factors

Features		Descriptive Statistics			Univariate Test
Factors	**Categories**	**Total N (%)**	**Negative N (%)**	**Positive N (%)**	χ^2 **(p-value)**
Smoking	No	492 (82.0)	356 (83.8)	136 (77.7)	0.08
	Yes	108 (18.0)	69 (16.2)	39 (22.3)	
Patients in Family	No	153 (25.5)	118 (27.8)	35 (20.0)	0.04
	Yes	447 (74.5)	307 (72.2)	140 (80.0)	
Number of Members with Complaint of Disease	0	153 (25.5)	118 (27.8)	35 (20.0)	0.18
	1	265 (44.2)	180 (42.4)	85 (48.6)	
	2	151 (25.2)	109 (25.6)	42 (24.0)	
	3	21 (3.5)	11 (2.6)	10 (5.7)	
	4	7 (1.2)	5 (1.2)	2 (1.1)	
	5	3 (0.5)	2 (0.5)	1 (0.6)	
Water Resources	mixed	105 (17.5)	73 (17.2)	32 (18.3)	0.79
	tape water	375 (62.5)	264 (62.1)	111 (63.4)	
	Well	120 (20.0)	88 (20.7)	32 (18.3)	
Socio-economic Status	High	31 (5.2)	20 (4.7)	11 (6.3)	0.52
	Low	80 (13.3)	60 (14.1)	20 (11.4)	
	Medium	489 (81.5)	345 (81.2)	144 (82.3)	
Toilet use	Combine	568 (94.7)	400 (94.1)	168 (96.0)	0.35
	Individual	32 (5.3)	25 (5.9)	7 (4.0)	
Awareness	No	503 (83.8)	359 (84.5)	144 (82.3)	0.51
	Yes	97 (16.2)	66 (15.5)	31 (17.7)	

Positive family member and district-wise distribution found significantly associated with seropositive subjects were analyzed further using multivariate analysis. A positive family member has 1.147 times higher odds of having of *H. pylori* positivity asymptomatically (OR: 1.147 CI 0.942 - 1.396). District wise distribution of *H. pylori* infection have presented the highest prevalence in Swat (50.3%), followed by Shangla (28.6%), Dir (6.9%), Buner (5.7%), Malakand (5.1%), and Kohistan (3.4%). The District-based

prevalence of *H. pylori* was found significantly associated in Swat but with lower odds (OR: 0.389 CI 0.213 - 0.714) (Table 3).

Table 3: Multivariate analysis of associated factors with seropositivity

Associated Factors		Descriptive Statistics		Multivariate Test	
Factors	**Categories**	**N**	**(%)**	**Odds ratio (CI)**	**p-value**
Positive family member	Positive	140	80.0	1.147 (0.942 - 1.396)	0.170
	Negative	35	20.0	**ref**	
District	Dir	12	6.9	0.867 (0.460 - 1.640)	0.658
	Kohistan	6	3.4	0.665 (0.299 - 1.447)	0.309
	Malakand	9	5.1	0.590 (0.279 - 1.224)	0.159
	Shangla	50	28.6	0.684 (0.361 - 1.300)	0.244
	Swat	88	50.3	0.389 (0.213 - 0.714)	0.002
	Buner	10	5.7	**ref**	

4. Discussion

The current investigation was aimed at finding the prevalence of seropositivity in asymptomatic individuals and their demographic risk indicators. A detection rate of 29.2% of seropositivity in our study is dissimilar to previous studies from Turkey (70%) [18], India (79%) [19], Libya (76%) [20], and Saudi Arabia (70%) [21], Brazil (65%) [22] and Bangladesh (90%) [23]. A higher infection rate in males in our studies is like previous studies where males have higher percentage of acquiring the infection [18,24,25]. There is no significant association of gender and age with serologically positive individuals. Similar results were stated in studies from Turkey and South Korea with higher frequency of males and age group >50 years but have no association with *H. pylori* positivity [18,24]. A higher infection rate in age group >50 years is parallel to studies previously reported where a low infection rate is observed in childhood and higher rate in adults [18,24,26]. A higher infection rate in adults is a trigger of childhood acquired infection that persists lifelong rather than increasing age as risk of infection.

A higher infection rate of 78.9% in married individuals in our study is suggestive of transmission between spouses. Previous study from Turkey has similar results with higher rates in married individuals [18]. Previous studies by Brenner in 1999 and 2006 [27,28] showed a higher percentage in individuals with infected partner. Although there is no significance of marital status on the *H. pylori* seropositivity.

Smokers have *H. pylori* seropositivity rate of 22.3% which agrees with previously reported study [18]. A recent study published in 2019 reported a greater risk of acquiring gastric cancer in seropositive smokers

[29]. An increased rate in uneducated people (39.4%) is comparable to a recent study in 2019 with higher rate (69.2%) among illiterate people [30]. A significant contrasting association was found in previous studies of infection with educational status [30,31]. A higher proportion of positive participants uses tape water (63.4%) which is consistent with previous study from UAE (58.6%), however dissimilar to our study, tape water users were significantly associated with infection in that study [32]. The only associated factor in our study from univariate analysis was positive family member which although is non-significant in regression model but has higher odds of having seropositivity. Family history with current or previous peptic ulcer was previously reported [32]. The possible cause of this association is the transmission route of the infection that can most efficiently be transmitted between family members or individuals sharing rooms and toilets.

Limitations of the Study

The limitations of the study are its limited number of participants. A large sample size would help in overcoming the shortcomings of the study in future.

Conclusion

The prevalence and contributing factors have been investigated for the first time among non-symptomatic individuals from Northern Province, KPK Pakistan. Among the included factors, the chance of infection is higher among individuals with positive family members. Understanding the infection rate in association with demographically associated factors will aid in managing the disease by adopting some prevention strategy.

Conflict of interest

The authors declare no conflict of interest.

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Author's Contribution

SARS.: conceptualization, designing, investigation and writing original draft. **SS.:** Validation and writing. **SB.:** formal analysis, data curation and writing. **AT.:** Analysis and validation.

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