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Helicobacter Pylori Infection: Epidemiological, Clinical and Pathological Aspects in a Digestive Endoscopy Unit and the Pathological Anatomy Service of Parakou in Benin Republic

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Abstract

Introduction: *Helicobacter pylori* (*Hp*) infection is the most common bacterial infection in the world. World Health Organization has classified this bacterium as a Class I carcinogen. The objective of this work is to study the epidemiological, clinical and pathological aspects of *Helicobacter pylori* infection among adults in a digestive endoscopy unit and the pathological anatomy service of Parakou. **Patients and Methods:** This was a descriptive and analytical cross-sectional study conducted from January 2020 to September 2020 in the digestive endoscopy unit and pathological anatomy service at the Departmental University Hospital Center of Borgou. Patients aged 18 years and older, who had undergone a gastroscopy with biopsies and who gave their oral consent were included. Three antral biopsies and two fundic biopsies were performed. The search for *Hp* was done under an optical microscope after staining with hematoxylin eosin. **Results:** A total of 151 patients were included and 51.66% were infected with *Hp*. Their average age was 40.63 ± 15.32 years and the sex ratio was 0.9. Epigastric pain was the most reported symptom (71.01%). Endoscopically, the prevalence of erythematous gastropathy was 98.72%. The study of chronic *Hp* gastritis according to Sydney system had shown signs of activity in 100% of patients with *Hp* and glandular atrophy in 83.33% of them. Intestinal metaplasia and dysplasia accounted for 24.35% and 29.48% of cases, respectively. Factors associated with *Hp* infection were: absence of proton pump inhibitor intake prior to exami-

nation, absence of handwashing with soap and water before eating, erythematous aspect of gastritis on gastroscopy, absence of intestinal metaplasia of the fundus, active and chronic aspects of gastritis on histology. **Conclusion:** The frequency of *Hp* infection in hospitals among adults admitted to the Digestive Endoscopy Unit of Parakou is high. To reduce this prevalence, it will be necessary to act on the factors identified.

Keywords

Helicobacter pylori, Gastroscopy, Pathological Anatomy, Parakou, Benin Republic

1. Introduction

Helicobacter pylori (*Hp*) is a micro-aerophilic and gram-negative spiral bacterium whose main reservoir is human stomach [1]. *Hp* infection is the most common bacterial infection in the world, affecting more than half of the world's population [2]. Its prevalence varies from country to country and depends on the level of socio-economic development [3]. According to a recent study, the prevalence of this infection is higher in Africa (79.1%), South America (69.4%) and Asia (54.7%) while it is lower in North America (37.1%) and Oceania (24.4%) [2]. In Benin, a seroprevalence of 75.4% in urban areas (Cotonou) and 72.3% in rural areas (Pahou) were found [4].

The risk of becoming infected with *Hp* changes depending on race or ethnicity, household characteristics, and geographic location [5]. Low socioeconomic conditions are associated with a high seroprevalence of *Hp* infection [6].

All routes of *Hp* contamination are still poorly understood. However, epidemiological studies recognize certain modes of transmission, the main one being human-to-human transmission by direct contact: oral-oral or fecal-oral [7]. Indirect contamination by water sources and food is also evoked as well as more rarely an iatrogenic pathway during digestive endoscopies [7].

Once contracted, *Hp* causes histologically chronic gastritis that persists throughout the life of the subject, but which remains most often asymptomatic [8]. In addition, it may be associated with a number of extradigestive manifestations [6]. Because of its involvement in pathologies such as gastric adenocarcinoma, extranodal lymphoma of the marginal areas annexed to the mucous membranes (MALT), and gastric atrophy, the World Health Organization (WHO) has classified it as a class I carcinogen [9].

Diagnostic methods for *Hp* infection are numerous. They can be invasive requiring gastric biopsy or non-invasive. Among these, we have: serology by ELISA, urea breath test and the search for bacterial antigens in stool [6]. Invasive tests include rapid urease test, pathological examination, culture and gene amplification [6].

The objective of this work was to study the epidemiological, clinical and pa-

thological aspects of *Hp* infection in adults admitted to a digestive endoscopy unit at Parakou, in the northern part of Benin.

2. Patients and Study Methods

2.1. Type and Period of Study

This was a cross-sectional study with descriptive and analytical purposes. Data collection covered the period from January 2020 to September 2020.

2.2. Study Population

The study population consisted of all patients who underwent gastroscopy in the digestive endoscopy unit at the Departmental University Hospital Center of Borgou (CHUD-B) during the data collection period.

2.3. Inclusion Criteria

The study included patients aged 18 years and older who performed a gastroscopy with gastric biopsies and gave their oral consent to participate in this study.

2.4. Exclusion Criteria

Patients who could not perform pathological examination of gastric biopsies or with blood dyscrasias precluding gastric biopsies were excluded from this study.

2.5. Variable

The dependent variable was the presence of *Hp* on histology. The independent variables were epidemiological, clinical and paraclinical data of patients.

2.6. Sampling

The sample size was calculated using Schwartz's formula [10]. With "N" the minimum number of subjects to be investigated; "z^2" confidence level = 1.96;

"p" prevalence (89.4%) [11]; "i" the margin of error = 5% and q = 1 − p =10.6%;

N = 146 people, in anticipation of 3% losses, N was increased to 146 + (3% of 146);

So the minimum sample size is 150 people.

Recruitment of patients meeting the inclusion criteria was comprehensive.

2.7. Medical Examinations Procedure

Gastroscopy was performed by the same hepato-gastroenterologist, without sedation, in patients who had been fasting for at least six (6) hours. Five (5) gastric biopsies were taken, including two antral at about 2 cm from the pylorus, two fundic, one at the angulus and put in different vials. The samples are immediately fixed with 10% neutral buffered formalin and then sent to the pathological anatomy laboratory with a form completed by the doctor.

It should be noted that in case of poor tolerance of gastroscopy, not all 5 biop-

sies were performed.

The histological examination of all samples was performed by the same pathologist assisted by two laboratory technicians. It took place in several stages including macroscopy, circulation, inclusion, microtomy, staining, assembly and microscopy.

At macroscopy, each biopsy was the subject of a macroscopic report that detailed the nature of fixative, number of fragments, dimensions, color and consistency of the biopsy. After measurement and description, the biopsy fragments were introduced into cassettes numbered according to their antral or fundic topography.

The circulation, consisted in making stay in a series of baths, the gastric biopsies put in cassettes, in order to give them rigidity by transforming them into a rigid firm tissue favoring the microtome cut. This circulation was done in three stages: 1) dehydration during which the tissue is cleared of the water it contains by following four baths of alcohol (ethanol) of increasing degree: 80˚, 90˚, 95˚ and 100˚ lasting one hour each; 2) lightening, which consisted in replacing ethanol in the tissue with xylene, and 3) impregnation, during which two paraffin baths of one and three hours respectively are performed.

Inclusion consisted of including the impregnated tissue in a block made of the inclusion medium (paraffin), melted between 55 and 60 degrees.

During microtomy, cuts of about 2 to 3 micrometers thick were made using a microtome (the Microtec CUT 4060).

The paraffin ribbons containing the biopsy tissue were spread on slides. The slides were stained with Hematoxylin and Eosin and then mounted with a special glue (Eukitt).

At microscopy and low magnification, the type of gastric mucosa (antral, fundic) was identified. At high magnification, it was specified the type of mucosa and the nature of different lesions (inflammation, activity, atrophy, intestinal metaplasia, dysplasia or even cancerization) as well as the presence or absence of *Hp*. The classification system for gastritis used in our study was that of Sydney.

2.8. Data Collection

Patient recruitment was done each time an upper GI endoscopy was performed. After obtaining their oral consent, patients selected for the survey were interviewed through a semi-structured interview. Paraclinical data were entered on the survey sheet as soon as the results were available.

2.9. Data Processing and Analysis

Data were recorded in Epi Data Entryclient 3.1 software. They were analyzed with Epi Data analysis 2.3 software. The significance threshold was set at 5%.

2.10. Ethical Considerations

Before this study, the protocol had obtained the agreement of the Local Ethics Committee for Biomedical Research of the University of Parakou (No.

0303/CLERB-UP/P/SP/R/SA). The patient anonymity was respected.

3. Results

3.1. Frequency of *Hp* Infection

Among the 151 patients included in the study, *Hp* was noted on histology in 78 subjects, representing a hospital frequency of 51.66%.

In 151 patients included, 146 had at least one antral biopsy and 140 had at least one fundic biopsy.

3.2. Characteristics of Patients Infected with *Hp*

- ***Socio-demographic data***

The mean age of patients infected with *Hp* was 40.63 ± 15.32 years with the extremes of 18 and 89 years. The most represented age group was 20 to 29 years (24.36%). Forty-one (52.56%) were female, or a sex ratio of 0.90. In 78 patients infected with *HP*, 46 (58.97%) were educated. The average household size was 5.97 ± 3.90 with the extremes of 1 and 15. More than half (53.85%) of subjects from households of more than 4 people were infected with *Hp*. The monthly income was lower than the guaranteed minimum interprofessional wage (SMIG) in Benin, *i.e.* less than 40,000 FCFA in 47 patients infected with *Hp* (60.26%).

- ***Lifestyle data***

Among the patients infected with *HP,* 42 (53.85%) reported daily use of potable drinking water (tap). Handwashing with soap and water was practiced by 43 people (55.13%) and after toilets by 50 people (64.10%). 63 people (80.77%) washed fruits and vegetables with simple water before consumption. In 78 patients infected with *HP*, 61 (78.21%) had latrines and 27 (34.61%) lived in mud or bamboo houses. Among the 78 subjects infected with *HP*, 75 people (96.15%) were non-smoking and 74 people (94.87%) did not take alcoholic beverages.

- ***Data on symptoms***

Sixty-nine (88.46%) patients infected with *Hp* had abdominal pain, located in the epigastric region in 71.01% of cases. Early satiety was reported by 55 patients (70.51%).

- ***Endoscopic data***

Endoscopically, 77 patients (98.72%) had erythematous gastropathy. Gastropathy was erosive or ulcerated in 15 people (19.23%). Gastric or duodenal ulcers were found in 6 subjects (7.69%).

- ***Histological data***

All the patients infected with *Hp* had active chronic gastritis. As for cell atrophy, it was present in 65 infected patients (83.33%). Chronic *Hp* gastritis was antral in 64 patients (82.05%). Table 1 shows the distribution of patients infected with *Hp* according to the histological aspects of gastric biopsies.

3.3. Factors Associated with *Hp* Infection

In bivariate analysis, no statistically significant association was noted between

Table 1. Distribution of *Hp* infected patients by histological aspects of gastric biopsies (Parakou, 2020, N = 78).

	Size	%
Active chronic gastritis		
Yes	78	100.00
No	0	00.00
Atrophic chronic gastritis		
Yes	65	83.33
No	13	16.67
Intestinal metaplasia		
Yes	19	24.35
No	59	75.65
Dysplasia		
Yes	23	29.48
No	55	70.51
Gastric cancer		
Yes	1	1.28
No	77	98.72

age (p = 0.830), sex (p = 0.571), socioeconomic status (p = 0.237), household size (p = 0.684), source of drinking water (p = 0.766), presence or absence of latrine at home (p = 0.691), type of dwelling (p = 0.163), alcohol (p = 0.292) or tobacco (p = 0.999) consumption and *Hp* infection (Table 2).

Failure to take a PPI two weeks before the examination (p = 0.040; RP = 0.68), absence of handwashing with soap and water before meals (p = 0.009; RP = 0.66), erythematosus aspect of gastritis (p = 0.023; RP = 0.37), absence of intestinal metaplasia in the fundus (p = 0.036; RP = 4.31) and active chronic gastritis (p = 0.005) were statistically associated with *Hp* infection, as shown in Table 3.

4. Discussion

This work has made it possible to have basic data on *Hp* infection in the northern part of Benin Republic. Moreover, the factors favoring this infection have been identified to better guide prevention.

The hospital frequency of *Hp* infection remains high in Parakou. It was 51.66%. It is close to 51.4% reported by Darré *et al*. [12] in 2010 in Togo. On the other hand, Doh *et al*. [13] in 2016 in Senegal and Essadik *et al*. [14] in 2011 in Morocco found a higher frequency of 75.6% and 69.2% respectively. The use in these studies of special straining including modified Giemsa which makes it possible to better visualize *Hp* and also the study population which included all age groups could explain this difference.

In this study, the mean age of infected patients was 40.63 ± 15.32 years. Doh

Table 2. Relationship between *Hp* infection and socio-demographic, economic, environmental and lifestyle characteristics of patients (Parakou, 2020, N = 151).

	Total (N)	*Hp* presence		PR	[95% CI; PR]	p
		n	%			
Age (years)						0.830
<20	7	5	71.43	1.29	0.61 - 2.72	
[20 - 29]	35	19	54.29	0.98	0.51 - 1.89	
[30 - 39]	36	17	47.22	0.85	0.43 - 1.68	
[40 - 49]	34	17	50.00	0.90	0.46 - 1.77	
[50 - 59]	17	10	58.82	1.06	0.52 - 2.15	
[60 - 69]	13	5	38.46	0.69	0.28 - 1.71	
≥70	9	5	55.56	1		
Sex						0.571
Male	75	37	49.33	1		
Female	76	41	53.95	1.09	0.80 - 1.49	
Level of Education						0.684
University	44	21	47.73	0.92	0.62 - 1.37	
Secondary	32	20	62.50	1.21	0.84 - 1.74	
Primary	13	5	38.46	0.75	0.36 - 1.54	
None	62	32	51.61	1		
Size of household						0.684
<4	48	26	54.17	1.25	0.73 - 2.12	
[4 - 10]	80	42	52.50	1.21	0.72 - 2.01	
>10	23	10	43.48	1		
Monthly income						0.237
<40,000 FCFA	84	47	55.95	1.21	0.88 - 1.67	
≥40,000 FCFA	67	31	46.27	1		
Source of drinking water						0.766
River	10	6	60.00	1.12	0.64 - 1.96	
Tap	85	42	49.41	0.92	0.67 - 1.28	
Well	56	30	53.57	1		
Existence of latrines						0.691
Yes	120	61	50.83	0.93	0.64 - 1.33	
No	31	17	54.84	1		
Type of inhabited house						0.163
Tiled	10	3	30.00	1		
In cement	98	48	48.98	1.63	0.62 - 4.30	
Mud house	28	19	67.86	2.26	0.85 - 6.03	

Continued

Bamboo house	15	8	53.33	1.78	0.62 - 5.12	
Smoking						0.999
Chewing tobacco	2	1	50.00	0.97	0.24 - 3.90	
Snuff	2	1	50.00	0.97	0.24 - 3.90	
Smoking tobacco	2	1	50.00	0.97	0.24 - 3.90	
Non-smoking	145	75	51.72	1		
Alcoholism						0.292
Yes	11	4	36.36	0.69	0.31 - 1.53	
No	140	74	52.86	1		

Table 3. Factors associated with *Hp* infection (Parakou, 2020, N = 151).

	Total (N)	*Hp* presence		PR	[95% CI, PR]	p
		n	%			
PPI intake before examination						0.040
Yes	44	17	38.64	0.68	0.45 - 1.02	
No	107	61	57.01	1		
Handwashing before meals						0.009
Yes	98	43	43.88	0.66	0.49 - 0.89	
No	53	35	66.04	1		
Erythematous gastritis						0.023
Yes	143	77	53.85	4.31		
No	8	1	12.50	1		
Intestinal metaplasia						0.036
Absent	130	71	54.62	1		
Present	10	2	20.00	0.37	0.11 - 0.99	
Active chronic gastritis						0.005
Yes	144	78	54.17			
No	7	0	0.00	1		

PPI = Proton Pump Inhibitor.

et al. [13] in 2016 in Senegal had found an average age close to ours (39.9 years). Our results are also similar to those of Itoudi *et al*. [15] in 2014 in Gabon and Hafidi *et al*. [16] in 2012 in Morocco, which noted a mean age of 41.9 years and 43.5 years respectively. The young age of infected patients confirms the fact that in developing countries, *Hp* infection is contracted at an early age. Indeed, a study conducted in the general population among children in the same city of Parakou reported a prevalence of 60.4% [17].

In this study, the majority of patients infected with *Hp* had abdominal pain (88.46%) and early satiety (70.51%). The site of abdominal pain was epigastric in

71.01% of cases. In the study conducted by Doh *et al*. [13] in 2016 in Senegal, epigastralgia (90.2%) was the most common sign. Hafidi *et al*. [16] in 2012 in Morocco, meanwhile, found epigastralgia in 65.81% of cases. Abdominal pain therefore remains the main clinical manifestation of *Hp* infection.

In patients with erythematous, erosive or ulcerated gastropathy, 53.85%, 63.16% and 50% respectively were infected with *Hp*. The same observation was made in Tunisia by Jmaa *et al*. [18] who reported in 2005 that 93.6% of patients who had gastropathy were infected with *Hp*. Gastropathy on gastrointestinal endoscopy would therefore be the most common elementary lesion of *Hp infection.*

Among cases of chronic *Hp* gastritis, atrophy was noted in 83.33% of cases. These data are lower than those of Essadik *et al*. [14] in 2011 in Morocco who reported a chronic atrophic gastritis prevalence of 91.8% in the presence of *Hp*. Darré *et al*. [12] in 2010 in Togo noted a higher prevalence of chronic *Hp* gastritis cases at 99%. This difference could be explained by the inclusion in our study of patients who took a PPI or antibiotics before biopsy, thus decreasing the possibilities of visualizing *Hp* on histology. Indeed, this bacterium induces inflammatory and immunological responses reflecting this significant proportion of atrophy in chronic *Hp* gastritis. This atrophy predisposes to metaplasia during which dysplasia or even cancer may appear [19].

In this study, living in a crowded household was not a risk factor for *Hp* infection ($p = 0.684$), whereas Ategbo *et al*. [20] in 2012 in Gabon and Agossou *et al*. [17] in 2018 in Benin, showed that overcrowding in households was a risk factor for *Hp* infection. This difference could be explained by the difference in the diagnostic method, in particular the detection of bacterial antigens in stool. These studies were also conducted during childhood, a period during which contamination would be greater.

This study found that taking a PPI during the last two weeks before gastric biopsy decreased the chances of visualizing *HP.* Indeed, patients who had not taken a PPI were more infected (57.01%) than those who had taken it (38.64%) and this difference was statistically significant ($p = 0.040$). This result could be explained by the inappropriate prescription of *Hp* eradication treatment observed in some of our patients. Indeed, most patients admitted for gastroscopy and from other hospitals had already received a PPI or even *Hp* eradication treatment without confirmation. This inappropriate prescription would be linked on the one hand to the limited financial means of patients to honor gastroscopy with biopsies for histopathological examination and on the other hand to the lack of technical platform necessary for the rapid diagnosis of *Hp* in all care facilities.

This work has shown that not washing hands with soap and water before meals is a risk factor for *Hp* infection. Patients who did not practice it were more infected (66.04%) and this association was statistically significant. Sagbo *et al.* [21] in 2018 in Benin reported that not washing hands with soap and water be-

fore meals was a risk factor for *Hp* infection. These results could be explained by the hand transmission of *Hp* infection.

As for endoscopic aspects, only patients with erythematous gastropathy were more likely to be infected with *Hp* (p = 0.023). Itoudi *et al.* [15] in 2014 in Gabon had noted that the presence of gastric endoscopic and duodenal lesions (gastropathy, gastric ulcer, duodenal ulcer) was statistically associated with *Hp* infection (p = 0.001). The inclusion in our study of patients who took antibiotics or a PPI before gastric biopsy could justify this difference.

Regarding pathological aspects, the presence of signs of cellular activity was significantly associated with *Hp infection* (p = 0.005). Our study also found an association between the absence of intestinal metaplasia in the fundus and the presence of *Hp*. Indeed, *Hp* was visualized in 54.62% of cases in the absence of intestinal metaplasia against 20% in the presence of intestinal metaplasia and this difference was statistically significant (p = 0.036). This result may justify the fact that the presence of intestinal metaplasia in the stomach is generally associated with low *Hp* density due to changes in mucus and gastric acidity [22].

One of the limitations of this study is that GIEMSA staining could not be used, which could underestimate the frequency of *Hp infection* in hospital.

5. Conclusion

In Parakou, in the northern part of the Republic of Benin, the hospital frequency of *Hp* infection is high. All age groups are concerned. These infected patients often report epigastric pain. Erythematous gastropathy is the lesion most found on digestive endoscopy. As for histopathology, *Hp* infection is responsible for chronic active gastritis often atrophic with sometimes intestinal metaplasia or even dysplasia. Factors associated with this infection are the lack of proton pump inhibitor intake during the last two weeks prior to upper GI endoscopy, lack of handwashing with soap and water before meals, erythematous aspect of gastropathy on digestive endoscopy, absence of intestinal metaplasia in the fundus, chronic and active characteristics of gastritis on histology. Raising public awareness on the manifestations of this infection and compliance with hygiene measures will prevent complications.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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