Benha Journal of Applied Sciences (BJAS) **print: ISSN 2356–9751**
Vol. (7) Issue (5) (2022), (179-186) **online: ISSN 2356–976x**
http://bjas.journals.ekb.eg

Detection of Antimicrobial Resistance Genes in Children Suffering from HelicobacterPylori Infection

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Abstract:

Background: Adult populations across the globe have an infection rate of H. pylori ranging from 20% to 90%. In Western communities, infection rates vary from 30 percent to 60 percent; in Asian nations and poor countries, the rates might be as high as 90 percent. H. pylori infection was found in the stomachs of 72.38 percent of Egyptian students. According to reports, the incidence of H. pylori infections in Egypt ranges from 70 to 88 percent in patients with chronic active HCV infection, depending on the type of infection. The bacteria is typically passed on via bloodlines and is most often picked up as a youngster. Chronic gastritis, peptic ulcer, and gastric cancer may all occur from a lack of therapy for gas trick colonisation. It is our goal to: Antimicrobial resistance genes (metronidazole, amoxicillin, clarithromycin) in children with Helicobacter pylori infection are the focus of this investigation. An Upper Gastrointestinal Endoscopy or gastroscopy was performed on 20 children aged less than 18 years old who complained of recurring stomach discomfort and were found to have a positive test for Helicobacter Pylori infection over the course of this prospective investigation. Banha University Hospital's department of paediatrics is where the research is taking place (Hepatology and Gastroenterology clinic). A structured interview was used to gather information on the patient's age, sex, gender identity, location, onset and duration of abdominal pain as well as any other symptoms that may be associated with H. pylori infection, such as epigastric pain and vomiting, dyspepsia, and gastrointestinal bleeding. Results: There were a total of 20 patients who were randomly chosen to participate in the research. Weight 42, Height 147.2 BMI 19, BMI (Centile) 56,36 and BMI (Control) 12.10 were the averages for the Control Group, whereas for the Experimental Group, the averages were 12.7 BMI (Centile), 45.5 BMI, Height 148.5 BMI (19.24 BMI) and 66.86 BMI (Centile). The percentage of (symptoms associated with H. pylori infection) and (symptoms in the case of the control group) the percentage of Abdominal pain before meal 100%, Abdominal pain relieved by food 0%, Pain relieved by antacid 80%, Night pain 70%, Nausea 80%, Vomiting 60%, Pain aggravated by food 100%, Pain relieved by belching 30%, Abdominal distension 20%, Anorexia & Weight loss 60%, Heart burn 80% Symptoms in the Experimental group include 100% of the subjects reporting abdominal pain before meals, 0% reporting relief from abdominal pain after meals, 100% reporting pain relief from antacids, 90% reporting night pain, 80% reporting nausea, 50% reporting vomiting, 100% reporting pain aggravated by food, 20% reporting pain relief from belching, 10% reporting abdominal distension, 50% reporting anorexia and weight loss, 70% reporting heart burn, 40% reporting chest pain, and 40% reporting GIT blee.

Keywords: Antimicrobial, Resistance Genes, Children, Helicobacter Pylori.

1. Introduction

Adult populations across the globe have an infection rate of H. pylori ranging from 20% to 90%. In Western communities, infection rates vary from 30 percent to 60 percent; in Asian nations and poor countries, the rates might be as high as 90 percent [1]. H. pylori infection was found in the stomachs of 72.38 percent of Egyptian students. According to reports, the incidence of H. pylori infections in Egypt ranges from 70 to 88 percent in patients with chronic active HCV infection, depending on the type of infection. Family members are the primary source of transmission for the bacteria, which is often acquired during infancy. Chronic gastritis, peptic ulcer, and gastric cancer may all occur from a lack of therapy for gas trick colonisation. It is estimated that humans and H. pylori have coexisted for at least 100,000 years. H. pylori has evolved a broad range of survival and adaptation tactics over its lengthy history in the unfavourable stomach habitat in humans. [4] H. pylori is a spiral-shaped member of the Helicobacteriaceae family. "Helix" and "bakterion," two Greek words meaning "spiral" or "coil" and "small staff," respectively, are the ancestors of Helicobacter. Hepatitis pylori's genus name is derived from two Greek words, "pyle" which means "gate" and "ourus," which refers to the pylorus area of the stomach. There is a significant prevalence of resistance to clarithromycin and metronidazole, which is linked to characteristics such as geographic location, sex (male or female), ethnicity, age, and active ulcer disease. It is known that antibiotic resistance is widespread in H. pylori. Antibiotic susceptibility testing was performed on a limited number of patients and on just a small number of strains in the majority of investigations. Large multicenter surveillance studies with uniform detection techniques should be preferred for obtaining antibiotic resistance rates in order to avoid the likelihood of antibiotic resistance prevalence in H. pylori being underestimated or overestimated. Information on the antimicrobial susceptibility of H. pylori isolates is valuable in predicting treatment success, since antimicrobial resistance of H. pylori isolates and poor compliance with medication regimens are two significant factors of treatment failure for H. pylori infection. Antimicrobial resistance in H. pylori has the potential to be a major public health issue due to the high frequency of infection and the devastating consequences that may result. In situations when antimicrobial treatment regimens fail to eradicate H., a 12fold rise in resistance to antimicrobial drugs such as clarithromycin has been recorded. Resistance to

antibiotics varies geographically and has been linked to the general public's use of specific antibiotics (e.g., clarithromycin for respiratory infections, metronidazole for parasite diseases and dental infections, tetracycline for respiratory and bowel diseases, amoxicillin for streptoccal pharyngitis, urinary tract infections). In the treatment of H. pylori, the combination of clarithromycin, metronidazole, amoxicillin, tetracycline, and rifampicin was used for triple combination therapy. The medication resistance is a problem. [11]

2. Method

Participants: This Prospective study was conducted on 20 children from both sexes, aged less than 18 years old with recurrent abdominal pain, dyspepsia, dysmotility or reflux dyspepsia that motive the performance of Upper Gastrointestinal Endoscopy or gastroscopy and diagnosed as positive H pylori infection, The study is conducted in department of pediatrics, Banha University Hospital (Hepatology and Gastroenterology clinic).

Methods: All included patients were subjected tofull history taking usingData was gathered by structured interview regarding age, sex, residence, onset, course and duration of abdominal pain, other associated symptoms stressing on symptoms associated with H. pylori infection e.g. epigastric pain, vomiting, dyspepsia, gastrointestinal bleeding, history of previous medications or previous treatment of H.pylori and family history.

The Inclusion criteria are Children aged less than 18 years and from Both sexes are included. But the Exclusion criteria are Patients who received non-steroidal anti-inflammatory drugs, as well as antibiotics, H2 receptors antagonists or proton pump inhibitors in the past four weeks prior to the study, Patients with medical comorbidities or any other major chronic disease, Children younger than older than 18 years and Any children whose parents refuse participating in the study.

- **Clinical investigations**

This Prospective study was conducted on 20 children from both sexes, aged less than 18 years old with recurrent abdominal pain, dyspepsia, dysmotility or reflux dyspepsia that motive the performance of Upper Gastrointestinal Endoscopy or gastroscopy and diagnosed as positive H pylori infection, The study is conducted in department of pediatrics, Banha University Hospital (Hepatology and Gastroenterology clinic) from 2018 to 2021 .

- **Body Mass Index (BMI) Body mass index (BMI):**was calculated as (weight in kg / height square in m2),(Normal BMI = 18.5–24.9, underweight = BMI<18.5 and Overweight BMI = 25–29).

BMI for patients <2 years assessed by using Egyptian growth charts (2006). BMI z-scores for patients <2 years was calculated using the (WHO) Child Growth Standards and Growth Reference data

- **Anthropometric measurements (Weight and height).**

-Weight was measured in kg (to the nearest 100 grams) using an electronic digital scale and its accuracy was periodically verified using reference weights.

-Height was measured in cm (measured to the nearest mm); children were measured on scales with height gauges, the subject standing with back against the gauge and feet on the weighing platform. Patients' height and weight for age percentiles were checked according to Egyptian growth curves (2002).

Laboratory investigations:

1. **Blood test**: Analysis of a blood sample may reveal evidence of an active or previous H. pylori infection in your body. However, breath and stool tests are better at detecting active H. pylori infections than is a blood test.
2. **Stool test**: A laboratory test called a stool antigen test looks for foreign proteins (antigens) associated with H. pylori infection in your stool. As with the breath test, PPIs and bismuth subsalicylate can affect the results of this test, so your doctor will ask you to stop taking them for two weeks before the test.
   - **Radiololgical investigations:**Abdominal Ultrasonography: real time Abdominal Ultrasonography was done for all the patients included in the study for evalution of:Liver to detect size ,site, tecture , border, reflectivity, homogenicity , periportal thickening , hepatic veins and pattern.Spleen: size, splenic vein diameter and collaterals Any focal lesion(s):number , site , size, shape, echogenecity
   - **Upper gastrointestinal tract endoscopyand four antral biopsies** will be obtained from each patient. One biopsy will be tested for rapid urease test, that was performed using rapid urease liquid test kit (Bussero, Milan, Italy) and the other three gastric biopsies will be stored in sterile physiological saline and kept at −70 °C until processed. DNA extraction used directly for detection of 16S rRNA, rdxA, Pbp1, 23S rRNA mutation gene using PCR assays
   - **Molecular detection of resistant genes** to metronidazole (rdx gene) and amoxicillin (pbp1A gene) will be carried out by conventional PCR followed by sequencing of PCR products. While detection of 23S rRNA gene conferring clarithromycin resistance will be carried out by real-time PCR.

Statistical analysis

Data management and statistical analysis were done using SPSS version 28 (IBM, Armonk, New York, United States). Quantitative data were assessed for normality using the Shapiro-Wilk test and direct data visualization methods. According to normality testing, numerical data were summarized as means and standard deviations or medians and ranges. Categorical data were summarized as numbers and percentages. Comparisons between positive and negative resistance genes were done using independent t-test for numerical variables and Fisher's exact test for categorical variables. All P-values were two-sided. P-values less than 0.05 were considered significant.

3. Results

Table (1) Demographic and general characteristics of the studied patients.

General characteristics		
Age (years)	Mean ±SD	12 ±2
	Range	7 - 13
Sex	Males n (%)	8 (40.0)
	Females n (%)	12 (60.0)
Residence	Rural n (%)	12 (60.0)
	Urban n (%)	8 (40.0)
Weight (kg)	Mean ±SD	41.3 ±8.8
	Range	24 - 55
Weight centile	Median (range)	37.5 (10 – 75.0)
Height (cm)	Mean ±SD	147.1 ±11.2
	Range	122 - 160
Height centile	Median (range)	37.5 (5 – 90)
BMI	Mean ±SD	18.86 ±1.77
	Range	16.1 – 22.6
BMI centile	Median (range)	65 (43.6 – 88.7)

BMI: Body mass index

The mean age of the studied patients was 12 ±2 years and ranged from 7–13 years. About two-thirds were females (60.0%). Also, the rural residence was reported in about two-thirds (60.0%). The mean weight was 41.3 ±8.8 kg and ranged from 24-55 kg, and the median weight centile was 37.5. The mean height was 147.1 ±11.2 cm and ranged from 122-160 cm, and the median height centile was 37.5. The mean BMI was 18.86 ±1.77 and ranged from 16.1-22.6, and the median BMI centile was 65.

Table (2) Main complaint of the studied patients.

	n (%)
Nausea	15 (75.0)
Vomiting	10 (50.0)
Abdominal distension	6 (30.0)
Heartburn	10 (50.0)
Anemia	10 (50.0)

The most frequent complaint was nausea (75%), followed by vomiting, heartburn, and anemia (50% for each). The least frequent complaint was abdominal distension (30%).

Table (3) Clinical presentation of the studied patients.

Reflux like dyspepsia	**n (%)**
Pain aggravated by food	20 (100.0)
Pain relieved by belching	6 (30.0)
Heartburn	10 (50.0)
Night pain	14 (70.0)
Vomiting	10 (50.0)
Red flags	
Anemia	10 (50.0)
GIT bleeding	0 (0.0)
Failure to thrive	8 (40.0)
High ESR	0 (0.0)
Bowel habits	
Constipation	6 (30.0)
Diarrhea	0 (0.0)

The presentation of the studied patients was classified according to reflux like dyspepsia, red flags, and bowel habits. Regarding reflux like dyspepsia, the most frequent presentation was pain aggravated by food (100%), night pain (70%), heartburn (50%), vomiting (50%), and pain relieved by belching (30%). Regarding red flags, the most frequent were anemia (50%) and failure to thrive (40%). No patients had GIT bleeding or high ESR. Regarding bowel habits, about one-third (30%) had constipation, and no diarrhea was reported.

Table (4) Laboratory findings of the studied patients.

Radiological & lab findings		
Hemoglobin (gm/dl)	Mean ±SD	11.2 ± 0.8
	Range	١٠ – 12.3
MCV (fl)	Mean ±SD	77 ±2.5
	Range	74 - 82
MCH (pg)	Mean ±SD	27.6 ±2.2
	Range	24 - 31
WBCs (×10^9/L)	Mean ±SD	8.3 ±1.4
	Range	5.8 - 10
Platelets (×10^9/L)	Mean ±SD	313.8 ±54.7
		240 - 380
High ESR	n (%)	0 (0.0)
Abnormal urine analysis & culture	n (%)	0 (0.0)
Abnormal stool analysis & culture	n (%)	0 (0.0)
Positive H.pylori stool antigen	20 (100%)	20 (100.0)
Positive rapid urease test	20 (100%)	20 (100.0)
Abnormal abdominal US	n (%)	0 (0.0)

All patients showed normal abdominal US. The mean hemoglobin was 11.2 ± 0.8 gm/dl and ranged from ١٠ – 12.3 gm/dl. MCV and MCH means were 77.1 ±2.5 fl and 27.6 ±2.2 pg, respectively. MCV ranged from 74 – 82 fl, while MCH ranged from 24 – 31 pg. The mean WBCs was 8.3 ±1.4 ×109/L and ranged from 5.8 – 10 ×109/L. The mean platelets was 313.8 ±54.7 ×109/L and ranged from 240 – 380 ×109/L. All patients showed normal urine and stool analysis & culture. In addition, all patients had positive H.pylori stool antigen and rapid urease test. No patients hadhigh ESR.

Table (5) Endoscopy findings of the studied patients.

		n (%)
Esophagus	Normal	20 (100.0)
Stomach	Nodularity only	3 (15.0)
	Hyperemia only	9 (45.0)
	Nodularity & hyperemia	6 (30.0)
	Watermelon appearance	2 (10.0)
Duodenum	Hyperemia	7 (35.0)

Regarding Endoscopic findings, all patients showed normal esophagus. Stomach findings were hyperemia (45%), nodularity and hyperemia (30%), nodularity only (15%), and watermelon appearance (10%). About one-third of the patients had duodenal hyperemia.

Table (6) Histopathology findings of the studied patients.

		n (%)
Stomach	Non-atrophic H.pylori-associated gastritis	3 (15.0)
Duodenum	Dudenitis	10 (50.0)
Activity of infection	Mild	7 (35.0)
	Moderate	7 (35.0)
	Severe	6 (30.0)
Severity of infection	Mild	7 (35.0)
	Moderate	7 (35.0)
	Severe	6 (30.0)

Only 15% of the studied patients had non-atrophic H.pylori-associated gastritis. Half of the patients had duodenitis (50%). The activity and severity of infection were mild (35%), moderate (35%), and severe (30%).

Table (7) Resistance genes in the studied patients.

		n (%)
Resistance gene for metronidazole (rdxA)	Positive	14 (70.0)
Resistance gene for clarithromycin (23srRNA)	Positive	5 (25.0)
Resistance gene for amoxicillin (pbp1a)	Positive	10 (50.0)

Seventy percent of the patients had a positive resistance gene for metronidazole (rdxA). One-quarter of the patients (25%) had positive resistance genes for clarithromycin (23srRNA), and half of the patients had positive resistance genes for amoxicillin (pbp1a).

4. Discussion

In terms of epidemiology, host response, clinical symptoms, associated disorders, diagnosis, and therapy, Helicobacter pylori infection in children varies from infection in adults [12]

Chronic Helicobacter pylori (H. pylori) infection is still widespread. This infection, which has been declining in many regions of the globe, remains a significant risk factor for peptic ulcer disease, stomach malignancy, and dyspepsia. Tests for H. pylori in patients with GERD, functional dyspepsia, non-steroidal anti-inflammatory medications, iron deficiency anaemia, or a higher risk of stomach cancer development are disputed [13].

The treatment of H. pylori in clinical practise is still a problem for doctors. Treatment and recovery from H. pylori-related disorders may be greatly improved if the correct medications are used in conjunction with them. Peptic ulcers, dyspepsia, and maybe stomach cancer can all be prevented by eradicating the H. pylori infection, provided it is treated early in its natural course. However, each complication has a different proportional risk reduction [14].

H. pylori is now treated empirically. As a consequence, establishing a treatment plan should aim to achieve a cure rate close to 100 percent With J.P. and Pajares JM. at their side In the present, eradication failure is alarming, since it is caused by an antibiotic resistant H. pylori strain that already existed or by the creation of an entirely new resistant strain from a previously vulnerable one. Efforts to rid the planet of antibiotic-resistant bacteria are inversely proportional to their success [15].

Compared to adults, the pathology seen in children is generally less severe as a result of the host immune response being down-regulated in the young. H. pylori infection may cause peptic ulcer disease, however this is more common in teens. It is not necessary to test and treat H. pylori if a patient has recurrent stomach discomfort. [16].

TolC similar genes (particularly hp0605, hp0971, hp1327 and hp1489) have been shown to be crucial in this organism for the upregulation of efflux pump activity and hence likely for MNZ resistance in later research in H. pylori. H. pylori's significant expression of hefA, which inhibits the accumulation of MNZ and is considered to be the first step in the development of MNZ resistance, has also been identified. Excess exposure to MNZ might lead to alterations in enzymes involved in MNZ reductive activation that result in decreased enzymatic activity [17].

The age range of the patients in our current research was 7–13 years, with a mean of 12 2 years. Two-thirds of those surveyed were women (60.0 percent). About two-thirds of respondents said that they lived in the country (60.0 percent). Between 24 to 55 kilogrammes, the weight varied from 41.3 8.8 kilogrammes, and the median weight centile was 37.5 kilogrammes. The median height centile was 37.5 centimetres, with a range of 122-160 centimetres. The mean height was 147.1 centimetres. The median BMI centile was 65, while the mean BMI was 18.86 1.77.

There were 60 patients in this research who were diagnosed with H. pylori infection using the fast urease test and verified using gastric biopsies (amplicon was discovered at 110 bp). Forty patients (66.7%) were men and 20 patients (33.3%) were females [18].

According to Alfizah's research, patients with resistant genotype strains had a median age of 10.1 3.9 years, which was close to but somewhat higher than the 9.3 3.2 years reported by another study in Spanish paediatric patients from 1999–2001.

According to previous studies, resistance-genetic MIC values were in the range of 0.16 to N 256mg/L. [19].

Nausea was the most common symptom in our research, with vomiting, heartburn, and anaemia following close after (50 percent for each). Abdominal distension was the least common complaint (30 percent).

Reflux symptoms such dyspepsia, red flags, and bowel habits were used to categorise the individuals in the research. If you have dyspepsia or reflux-like symptoms, the most common ones you report are pain that is made worse by eating, nighttime discomfort, acid reflux, nausea, and pain that is reduced by belching (30 percent). More than half (50 percent) of all warning flags were anaemia and failure to thrive (40 percent). There were no reports of GIT bleeding or elevated ESR in any of the individuals. Constipation was reported by around one-third of the participants (30 percent).

According to another research, in the control group, the proportion of history of H pylori treatment is 100%, the history of prior medication is 0%, and family history of H pylori infection is 30%. There is a strong correlation between the two. In the Experimental group, the rate of prior H pylori treatment is 100%, previous medication history is 0%, and family history of H pylori infection is 40%. [20].

91.4 percent of patients investigated by J. P. Gisbert and X. Calvet had H. pylori isolates. Patients with poor socioeconomic position are more likely to get H. pylori infection. 65.5 percent of H. pylori-infected individuals also have a family history of the disease [20].

Researchers Nicoline F. Tanih and Roland N. Ndip found that all fifteen patients tested positive for primary clarithromycin resistance (100 percent) in the absence of any prior history of triple treatment. Similarly, Tüzün et al. (southeastern Anatolia) observed a similar finding. As for primary clarithromycin-resistant infections in Tunisia, the incidence was only 15.4%. The H. pylori isolates from previously treated patients were shown to be more resistant to clarithromycin than H. pylori strains isolated from patients who had not had any therapy by Acute and his colleagues, in contrast to our findings (583%) and 21%, respectively, in their analyses [21].

Abdominal X-rays were normal in all individuals. Hemoglobin levels varied from 10 to 12.3 gm/dl, with an average of 11.2 0.8 gm/dl. There were 77.1 2.5 fl in MCV and 27.6 2.2 pg in MCH. There were 8.3 1.4 WBCs/L, with a range of 5.8 – 10 109/L, in the WBC

count. The platelet count varied from 240 to 380 109/L, with a median of 313.8 54.7 109/L. Analysis and culture results for all patients were normal. The H. pylori stool antigen as well as the fast urease test were both positive in all of the patients. There were no cases of elevated ESR in any of the individuals.

All patients had normal esophageal endoscopic results. 45% of patients had hyperemia, 30% had both nodularity and hyperemia, 15% had just nodularity, and a watermelon look was seen in the stomach (10 percent). The duodenum was hyperemic in almost one-third of individuals.

Motamed et colleagues showed that nodularity was identified in 47.4% of H.pylori positive individuals, however this was about the same for mucosal erythema (24.8%), erosion and ulceration (5%), and normal stomach endoscopic findings (1.1%), which were lower than these findings [22].

A study of 49 children by Mrad et al indicated that H.pylori infection was identified in 35 of the children (71.5 percent). There was nodular gastritis seen in 16 of the 49 children studied, with 14 of 35 (40 percent) infected children and two of 14 (14 percent) non-infected children (p=0.07) showing signs of the condition [23].

174 children with helicobacter pylori infection had greater levels of stomach inflammation, according to a research by Luzza et al. (2001).

Eighty-four (48% of the children examined) showed signs of H. pylori infection, including nodularity in 40% of those tested positive, erythema in 13%, erosion and ulcer in 1% [24].

Non-atrophic H. pylori-associated gastritis was seen in only 15% of the patients in our research. Duodenitis affected half of the individuals (50 percent). (35 percent), moderate (35%), and severe (35%) activity and severity of infection were observed (30 percent).

Endoscopic evaluation by Motamed et al.,(2014) revealed that the majority of patients had a normal duodenum. There was duodenitis (the most prevalent pathology finding) in 6.4 percent of H.pylori positive individuals [22].

10.7 percent of H.pylori-positive patients showed some degree of duodenal inflammation, according to Luzza's et al. (2001) [24].

Of the 75 H.pylori positive children studied, 32/75 (42.7%) had mild H.pylori-associated gastritis and duodenitis, 37/75 (49.3%) had moderate to marked H. pylori density, and 6/75 (8%) had severe activity and H.pylori density with superifial ulceration in the gastric and duodenal mucosas.

Cardenas-Mondragon et al, who studied 333 paediatric patients with chronic abdominal pain, found that none of the H.pylori positive patients had normal gastric pathology findings, 31 percent of the total had mild gastritis, 14.9 percent of the total had moderately active gastritis, and 4% had severe gastritis in evaluation of biopsy specimens [25].

Helicobacter pylori gastritis in children has been researched by Galos et al. (2018) who found that 14/28 (50 percent) had mild H. pylori density, 11 (38.29 percent) had moderate density, and 3 (10.71 percent) of the 28 patients had notable density, according to their findings [26].

Gastritis and doudenitis caused by H. pylori are caused by adhesins (HopQ, HopP, and HopS) that allow the bacterium to adhere to the gastric epithelial cells in a receptor-specific manner, leading to the expression of virulence factors such as CagA and VacA, which are responsible for the bacterium's cytotoxic effects [27].

70 percent of the patients had a positive resistance gene for metronidazole, 25 percent had a positive resistance gene for clarithromycin, and 50 percent had a positive resistance gene for amoxicillin, according to our research (pbp1a).

Metronidazole (25 percent) and amoxicillin (18.3 percent) resistance rates were found in gastric biopsy specimens in Diab M etal, (2018) research, whereas the resistance rate to clarithromycin (6.7 percent) was found in gastric biopsy specimens utilising real time PCR for 23S rRNA detection.

Clarithromycin, a crucial component of traditional triple treatment for H. pylori, is the primary source of concern (18). Gisbert and others [29].

More H. pylori stomach samples in the present research had the A2143G mutation site linked with clarithromycin resistance than in the earlier Egyptian study (4 percent) by Sherif et al [30].

H. pylori's resistance to clarithromycin differed considerably across three geographic regions (17.5 percent in Europe, 18.9 percent in Asia and 29.3 percent in America). According to De Francesco et al. [31] and Liu et al., China and Japan had the highest rates of resistance in Asia at 84.9 percent and 40.7 percent, respectively [32].

Accordingly, it is essential to assess the prevalence of clarithromycin resistance rates in each geographic location so that treatment regimens may be better tailored to local conditions. Thung and others [33].

It's possible that our findings are a little off since we used various equipment and strains of bacteria. The PCR-RFLP approach was used by A. B. Olokoba et al. to look for mutations in the 23S rRNA gene in H. pylori isolates. That analysis found that the A2143G mutation was present in all of the resistant isolates. There is some evidence that stool-based detection is less sensitive than endoscopy, although it has the benefit of not necessitating endoscopy. Etest discovered a single resistant clone in both participants with a MIC of 1. mg/liter, although only wild-type alleles were found in ddPCR. [35].

An A2142C, G, or A2143G variant in the 23S rRNA gene (said to account for around 90% of all clarithromycin resistance) may be detected using our ddPCR test. The T2182C mutation, previously linked to poor resistance, was found in the sequences of the two weakly resistant isolates. While a broader range of mutations might be investigated in order to increase the accuracy of the ddPCR test, it is yet unclear if this mutation and low-level resistance are relevant to therapeutic failure [36].

Because we used various equipment and standard strains, the little variance in our findings (57.6oC) might be attributable to this. A. B. Izabela et al. used the PCR-restriction fragment length polymorphism (RFLP) approach to study 23S rRNA gene alterations in H. pylori isolates. That analysis found that the A2143G mutation was present in all of the resistant isolates. A-to-G or A-to-C point mutations at positions 2142 and 2143 were considered clinically significant in the research by Izabela et al, whereas other variants were deemed clinically insignificant. Both the US and Europe have a high prevalence of the A2143G and A2144G mutations seen in clarithromycin-resistant H. pylori when employing the PCR-RFLP technique. [37]

4. Conclusion

- Chronic and severe peptic ulcer disease caused by Helicobacter pylori infection is exceedingly frequent and was previously linked to stress, heredity and personality.
- As time went on, the frequency of primary resistance to clarithromycin, metronidazole and levo- floxacin remained high and grew, but the resistance rates to amoxicillin, tetracycline and furazoli-done remained low and steady.

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