Sains Malaysiana 48(12)(2019): 2675–2682
http://dx.doi.org/10.17576/jsm-2019-4812-08

Helicobacter pylori Infection: Prevalence, Demographic Characteristics, Clarithromycin Resistance and Evaluation of the In-House Rapid Urease Test in Sungai Buloh Hospital, Malaysia

(Jangkitan *Helicobacter pylori*: Prevalens, Ciri Demografi, Kerintangan Klaritromisin dan Penilaian Ujian Pantas Urease Dalaman di Hospital Sungai Buloh, Malaysia)

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ABSTRACT

Helicobacter pylori *infection remains an essential global research focus, and increasing clarithromycin resistance was reported to impact the efficacy of clarithromycin-based treatment regiments. The study objectives sought to understand the prevalence of* H. pylori *infection amongst patients from Sungai Buloh Hospital, Malaysia, its association with demographic factors, and the clarithromycin resistance rate. The in-house rapid urease test (IRUT) was also evaluated and compared to the* Campylobacter*-like organism (CLO) test using culture or histopathological testing as the gold standard for diagnosis. The gastric corpus biopsies of 352 patients were included, as well as their age group, gender and ethnicity. Clarithromycin susceptibility was measured using the E-test method. The overall prevalence of* H. pylori *infection was 15.1% (53/352). There was no significant association between the age groups, gender and ethnicity with regards to* H. pylori *infection. Four of the 13 viable isolates (30.8%) were clarithromycin-resistant. Although IRUT had a slightly lower specificity (94.9%) than that of the CLO test (95.9%), both tests had the same sensitivity values (81.1%). IRUT had a lower positive predictive value (74.1%) than the CLO test (78.2%) but showed a similar negative predictive value (96.5%) compared to the CLO test (96.6%). Both tests displayed a very good agreement (κ = 0.97). In conclusion, the overall prevalence of* H. pylori *infection in our study was generally low. The high proportion of clarithromycin-resistant isolates may not reflect the exact resistance rate due to the small number of positive cultures. Our IRUT is an acceptable alternative to the CLO test for the rapid diagnosis of* H. pylori *infections based on its comparable performance.*

Keywords: Clarithromycin; Helicobacter pylori*; rapid urease test*

ABSTRAK

Jangkitan Helicobacter pylori *kekal menjadi fokus penyelidikan global yang penting dan tahap kerintangan klaritromisin yang dilaporkan meningkat boleh memberi impak terhadap keberkesanan rejimen rawatan berasaskan klaritromisin. Objektif kajian ini adalah untuk mengkaji prevalens, sekutuan antara faktor demografi dan kadar kerintangan terhadap klaritromisin bagi jangkitan* H. pylori *dalam kalangan pesakit Hospital Sungai Buloh, Malaysia. Kami juga telah menilai dan membandingkan prestasi ujian pantas urease dalaman (IRUT) dengan ujian organisma seperti* Campylobacter *(CLO) menggunakan ujian kultur atau histopatologi sebagai piawai rujukan untuk diagnosis. Biopsi korpus gastrik daripada 352 pesakit telah dimasukkan dalam kajian ini dan data merangkumi kumpulan umur, jantina dan bangsa telah dianalisis. Kerentanan klaritromisin diukur menggunakan ujian E-test. Prevalens keseluruhan jangkitan* H. pylori *adalah 15.1% (53/352). Tiada sekutuan signifikan didapati antara jangkitan dan faktor demografi yang dikaji. Empat daripada 13 pencilan berdaya hidup (30.8%) adalah rintang klaritromisin. Walaupun ujian IRUT mempunyai kekhususan yang sedikit lebih rendah (94.9%) daripada ujian CLO (95.9%), namun kesensitifan kedua-dua ujian adalah sama (81.1%). Nilai ramalan positif ujian IRUT adalah lebih rendah (74.1%) berbanding ujian CLO (78.2%) namun nilai ramalan negatif hampir sama (96.5%) dengan ujian CLO (96.6%). Kedua-dua ujian menunjukkan persetujuan yang sangat baik (κ = 0.97). Kesimpulannya, prevalens keseluruhan jangkitan* H. pylori *dalam kajian kami pada umumnya adalah rendah. Kadar bilangan pencilan rintang klaritromisin yang tinggi mungkin tidak mencerminkan kadar rintangan sebenar memandangkan bilangan kultur positif yang sangat rendah. Prestasi ujian IRUT kami setanding dengan ujian CLO menjadikannya ujian alternatif yang boleh diterima sebagai ujian pantas bagi mengesan jangkitan* H. pylori.

Kata kunci: Helicobacter pylori*; klaritromisin; ujian pantas urease*

INTRODUCTION

Helicobacter pylori infects half of the world population with a much higher prevalence reported amongst developing countries. It is defined as a fastidious, microaerophilic, catalase and oxidase-positive, urease producing, spiral shaped gram-negative bacterium, which grows in a

relatively narrow pH range of 5.5 to 8.0 (Scott et al. 2002). *H. pylori* infection is the leading cause of peptic ulcer disease, which accounts for 90% of duodenal ulcers, and about 50 to 80% of gastric ulcers (Ernst & Gold 2000). It is identified as the major known risk factor associated with the development of gastric cancer (Suerbaum & Michetti 2002). Studies have reported a steady worldwide decline in the success rate of clarithromycin-based triple therapy for the eradication for *H. pylori* infection, primarily due to clarithromycin resistance, which underlines the importance of continued surveillance (Graham & Fischbach 2010; Mégraud 2004). In Malaysia, distinctive differences have been observed in the distribution of *H. pylori* infection amongst the different ethnicities living together in this country, consisting of Malays, Chinese and Indians (Goh 2009).

The diagnosis of *H. pylori* infection in Sungai Buloh Hospital is dependent on a commercially available rapid urease test, culture and histopathological examination (HPE) of gastric biopsies from endoscoped patients. The rapid urease test is the method of choice for indirectly detecting the presence of *H. pylori* in gastric biopsy specimens. It is easily performed, reliable and less time consuming in comparison to culture and histology methods (Roma-Giannikou et al. 2010) One of the most commonly used commercial rapid urease test with a high sensitivity, and specificity, is the Campylobacter-like organism test, or CLO (*Ballard Medical Products*, Draper, Utah, USA). However, it is relatively more expensive (Graham 1991). Among all these tests, a positive culture is deemed to have 100% specificity (van der Hulst et al. 1996) and remains the gold standard for *H. pylori* diagnosis. However, culture is time consuming, and requires handling by skilled personnel.

In this study, we sought to assess the prevalence, and association with demographical characteristics, as well as the clarithromycin resistance among *H. pylori* infections in Sungai Buloh Hospital. It is a 620-bedded tertiary hospital in the state of Selangor, Malaysia. It serves the districts of Gombak, Petaling and Kuala Selangor. This consists of a combined population of more than 2.8 million people, and is one of the major referral centres in Malaysia with an upper gastrointestinal subspecialty. We also produced our own in-house rapid urease test (IRUT) as per MacFaddin (1980), and conducted an evaluation and comparison of its performance using the CLO test.

Materials and Methods

Study Design and Population

This is a cross-sectional study conducted between August 2015, and May 2016, which involved patients who underwent elective endoscopy of the upper gastrointestinal tract at the surgical day care unit of Sungai Buloh Hospital, Malaysia. The eligibility criteria included patients aged 18 years and above who were suspected of contracting *H. pylor*i infection, as well as those who were not receiving amoxicillin/clavulanate, clarithromycin and levofloxacin 4 weeks prior to endoscopy. Patients who were currently, or previously taking proton pump inhibitors such as omeprazole, lansoprazole and esomeprazole within 10 to 14 days prior to endoscopy, were also excluded. However, patients taking pantoprazole were included, as its usage has been proven not to inhibit growth and the urease activity of *H. pylori* (Saniee et al. 2015).

Specimen Collection

For each eligible consented patient, four tissue biopsy specimens ranging from 0.1 to 0.5 mm in diameter were taken from the gastric corpus during elective endoscopy by the attending surgeon, according to the Hospital's standard practice. Two of the biopsy specimens were then immediately tested in the endoscopy room with the IRUT and CLO tests, respectively. One biopsy specimen was placed in a sterile container containing 1 mL of sterile physiological saline (0.9% NaCl), and transported at room temperature to the microbiology laboratory of Sungai Buloh Hospital, within 1 h from the time of collection. The specimen was then processed for culture in the microbiology laboratory within an hour of it being received. The remaining specimen was sent in formalin to the histopathology laboratory at Universiti Teknologi MARA (UiTM) for HPE.

Identification of Isolates

In-house rapid urease test The urea medium was prepared by adding and mixing 50 gm of urea broth with 500 mL of distilled water, and 8 mL of 1% phenol red, which acted as a pH indicator. The medium was tested with a pH meter, and with the aim of having a pH of 5.5. This was achieved using pH buffers in order to prevent the potential activity of urease-producing contaminating bacteria from producing enough ammonia to turn the colour of the pH indicator to pink, or magenta, within a specified period. The urea medium was stored at -20°C until further use.

For each test, 0.5 mL of prepared urea medium was distributed into sterile bullet tubes and stored in a dedicated chiller between 2-8°C. Quality control testing was done for every newly prepared batch using *Proteus mirabilis* ATCC® 43071 as the positive control group. A positive result was determined through observing colour change from yellow to pink, or magenta, over a period of 20 min (MacFaddin 1980).

CLO test The CLO test consisted of a well of urease indicator gel sealed inside a plastic slide, which contained urea, 29 mg/mL USP, phenol red as a pH indicator, buffers, and a bacteriostatic agent. The test was performed according to the manufacturer's instructions. A positive result was determined through colour change from yellow to bright magenta within 20 min.

Culture and Sensitivity The biopsy specimens were seeded in a Columbia horse blood agar (BA) plate containing a

Dent supplement, and were incubated at 37°C for up to 6 days in a sealed jar under microaerobic atmosphere using a CampyGen GasPak (Oxoid, Hampshire, United Kingdom). The plates were examined for growth every 2 days from the start of the incubation, up to the 6th day of incubation. *H. pylori* colonies tend to appear as small, grey, and translucent, with the presence of weak beta-haemolysis. The colonies were subsequently subjected to Gram stain, and were further tested with catalase, oxidase and the IRUT to facilitate its identification.

For clarithromycin susceptibility testing, the colonies were further sub-cultured onto 3 more BA plates to produce enough growth to meet the 2.0 McFarland colony inoculum standard as required for the minimum inhibitory concentration (MIC) testing. An E-test clarithromycin strip was placed onto a Mueller-Hinton blood agar which was inoculated with the colony inoculum, and was incubated at 37°C under microaerobic atmosphere using a CampyGen GasPak. The MIC was then read after 72 h of incubation. The interpretation was made according to the clarithromycin susceptibility breakpoints, as published by the Clinical and Laboratory Standards Institute (CLSI 2010).

Histopathological examination The biopsy specimens sent to the histopathology laboratory in UiTM were processed and stained with hematoxylin and eosin (H&E), to directly identify *H. pylori on* the gastric epithelium under high magnification light microscopy. Giemsa staining was also performed on all biopsy specimens to increase the sensitivity and specificity of the *H. pylori* detection.

H. pylori infection was diagnosed through either a positive culture, or a positive HPE, irrespective of the rapid urease test results.

DATA COLLECTION

Demographic data consisting of the patient's age, gender and ethnicity were collected via a data collection sheet by the attending surgeon performing the endoscopy procedure. The diagnosis and indication for endoscopy were also elicited for all patients. The HPE results from UiTM were retrieved via the electronic information system.

STATISTICAL ANALYSIS

Statistical analysis was performed with the Statistical Package for Social Sciences (SPSS) application for Windows version 21. Bivariate analysis was performed using the Pearson's Chi-Square and Fisher Exact test, to determine the association of the demographic data with the *H. pylori* infection. A p-value of <0.05 was considered as significant. The sensitivity, specificity, positive predictive value (PPV) and negative predictive value (NPV) (with 95% confidence intervals) of the IRUT and CLO tests were calculated via VassarStats (*Http://vassarstats.net/clin1.html)*. The strength of agreement between IRUT, CLO test, culture and HPE was also analysed using Cohen's Kappa. The Kappa value (κ) was interpreted according to Altman (1991).

RESULTS

The total number of patients included in this study was 352. All gastric corpus biopsy specimens received from the 352 patients were subjected to IRUT, CLO test, culture and HPE, except for 11 cases of which we did not receive biopsy specimens for culture testing.

DEMOGRAPHIC AND CLINICAL CHARACTERISTICS OF PATIENTS ENROLLED IN THE STUDY

The mean age of 352 patients included in the study was 52 (S.D. 16) years, with the age ranging from 18 to 92 years. Patients were categorised by age into young adults (ages 18 to 44 years), middle-aged adults (45 to 65 years) and the elderly (more than 65 years). The majority of the patients included were from the middle-aged group, followed by young adults and the elderly. Slightly more than half of the included patients were male (55.4%, 195/352). The majority of the patients were of Malay ethnicity (60.8%, 214/352), followed by Indians (19.6%, 69/352) and Chinese (17.9%, 63/352) (Table 1).

Slightly more than half of the included patients were diagnosed with acute or chronic gastritis (54.3%, 191/352) followed by peptic ulcer disease (22.2%, 78/352) and gastroesophageal reflux (7.9%, 28/352). In the minority cases, endoscopy was facilitated to rule out gastric

TABLE 1. Demographic data of patients enrolled in the study (N=352)

	Demographic factors	n (%)
Age groups	18 - 44 years	135 (38.4)
	45 - 65 years	150 (42.6)
	> 65 years	67 (19.0)
Gender	Male	195 (55.4)
	Female	157 (44.6)
Ethnicity	Malay	214 (60.8)
	Indian	69 (19.6)
	Chinese	63 (17.9)
	Others	6 (1.7)

malignancy (5.7%, 20/352), hiatus hernia (3.9%, 14/352) and gastrointestinal bleeding (3.1%, 11/352). Other less common indications included investigation for portal hypertensive gastropathy and anaemia (2.8%, 10/352).

OVERALL PREVALENCE OF *H. pylori* INFECTION IN SUNGAI BULOH HOSPITAL

The overall prevalence of *H. pylori* infection in Sungai Buloh Hospital was 15.1% (53/352). Out of the 53 positive cases, 12 were diagnosed with both a positive culture and HPE, whereas 39 were diagnosed with HPE only, and 2 with culture only (Table 2).

ASSOCIATION BETWEEN DEMOGRAPHIC FACTORS AND *H. pylori* INFECTION

The mean age of 53 patients with *H. pylor*i infection was 49 (S.D. 15) years, with the age ranging from 22 to 85 years. There was a higher proportion of positive cases among the young adult age group (17.8%, 24/135), male patients (16.4%, 32/195) and Indian ethnicity (18.8%, 13/69). However, there was no statistically significant association between the age group, gender and ethnicity with *H. pylori* infection (Table 3).

SENSITIVITY, SPECIFICITY, POSITIVE AND NEGATIVE PREDICTIVE VALUE OF IN-HOUSE RAPID UREASE TEST AND CLO TEST COMPARED TO THE GOLD STANDARD

The diagnostic accuracy of the IRUT compared to that of the culture or HPE, which serves as the gold standard, showed a sensitivity and specificity of 81.1%, and 94.9%, respectively. The CLO test showed a sensitivity of 81.1%, which was equivalent to the IRUT. However, it showed a slightly higher specificity of 95.9% (Table 4). The PPV and NPV for IRUT was 74.1% and 96.5%, respectively. The CLO test had a higher PPV of 78.2%, and a similar NPV of 96.6%, compared to the IRUT (Table 5).

AGREEMENT BETWEEN IN-HOUSE RAPID UREASE TEST WITH CLO TEST, CULTURE AND HISTOPATHOLOGICAL EXAMINATION

The strength of agreement was very good between IRUT and the CLO test ($\kappa = 0.97$), good between IRUT and HPE ($\kappa = 0.73$), and fair between IRUT and culture ($\kappa = 0.27$).

CLARITHROMYCIN RESISTANCE

Out of 53 cases diagnosed with *H. pylori* infection, only 14 specimens from the cases were obtained from the culture-positive, and were subjected to clarithromycin

TABLE 2. Culture, histopathological examination, in-house rapid urease test and CLO test results of the 53 *H. pylori*-positive cases

Culture	HPE	IRUT	CLO test	n
+	+	+	+	10
-	+	+	+	32
+	-	+	+	1
+	+	-	-	2
+	-	-	-	1
-	+	-	-	7

+ : Positive, - : Negative

TABLE 3. Association between *H. pylori* infection and demographic factors (N= 352)

Demographic factors			*H. pylori* positive	*H. pylori* negative	
		Total	n (%)	n (%)	*p*-value
Age groups	18-44 years	135	24 (17.8)	111 (82.2)	0.254[a]
	45-65 years	150	23 (15.3)	127 (84.7)	
	>65 years	67	6 (9)	61 (91)	
Gender	Male	195	32 (16.4)	163 (83.6)	0.429[b]
	Female	157	21 (13.4)	136 (86.6)	
Ethnicity	Malay	214	29 (13.6)	185 (86.4)	0.492[c]
	Chinese	63	10 (15.9)	53 (84.1)	
	Indian	69	13 (18.8)	56 (81.2)	
	Others	6	1 (16.7)	5 (83.3)	

a- Pearson's Chi-Square, $X^2 - 0.254$ (df 1)
b- Pearson's Chi-Square, $X^2 - 0.626$ (df 1)
c- Fisher Exact test

TABLE 4. Results of in-house rapid urease test and CLO test compared to culture or histopathological examination as the gold standard to detect *H. pylori*

		H. pylori detection with the gold standard		
Test	Result	Positive n (%)	Negative n (%)	*p*-value
IRUT	Positive	43 (81.1)	15 (5.1)	<0.001[a]
	Negative	10 (18.9)	284 (94.9)	
CLO test	Positive	43 (81.1)	12 (4.1)	<0.001[b]
	Negative	10 (18.9)	287 (95.9)	

a- Pearson Chi square, X^2 – 189.5 (df 1)
b- Pearson Chi square, X^2 –230 (df 1)

TABLE 5. Sensitivity, specificity, positive predictive value and negative predictive values with 95% CI for in-house rapid urease test and CLO test

Test	Sensitivity (%)(95% CI)	Specificity (%)(95% CI)	PPV (%)(95% CI)	NPV (%)(95% CI)
IRUT	81.1 (67.5-90.1)	94.9 (91.6-97.0)	74.1 (60.7-84.3)	96.5 (93.6-98.2)
CLO test	81.1 (67.5-90.1)	95.9 (92.9-97.8)	78.2 (64.6-87.8)	96.6 (93.7-98.3)

PPV = positive predictive value; NPV = negative predictive value; CI = confidence interval

susceptibility testing. From the 14 isolates, 1 was non-viable for testing, 9 isolates were susceptible to clarithromycin with an MIC of ≤ 0.25 μg/mL, and 4 isolates were clarithromycin-resistant with a MIC of ≥1 μg/mL.

Discussion

The overall prevalence of *H. pylori* infection in Sungai Buloh Hospital (15.1%) was generally low, and was comparable with local studies conducted in North Eastern Peninsular Malaysia (13.5%) (Kaur & Naing 2003), and in Serdang Hospital (15.6%) (Chieng et al. 2015). The majority of the study population in both of these studies (and in ours) comprised of Malays. In contrast, a higher overall prevalence of 30.8% was recorded in Universiti Kebangsaan Malaysia Hospital, whereby the ethnic majority of the study population was Chinese (Alfizah et al. 2010). In addition, larger multicentre seroepidemiologic studies have recorded much larger variations with respect to the overall prevalence of different geographical areas across Malaysia. These seroprevalence rates ranged between 26% and 55% (Goh & Parasakhti 2001). Worldwide, an overall prevalence of *H. pylori* infection varies from region to region, with the prevalence ranging from 18.9% in Switzerland, to 87.7% in Nigeria (Hooi et al. 2017).

Age did not prove to be a significant factor for *H. pylori* infection in our study, which was consistent with the study in North Eastern Peninsular Malaysia (Kaur & Naing 2003). A different study showed an association between age and *H. pylori* infection, with the mean age of patients diagnosed with *H. pylori* infection being much older (53.99, S.D. 14.74), compared to those without the infection (51.99, S.D. 15.82) (Alfizah et al. 2010). In contrast, another local study in Sungai Petani Hospital recorded a higher prevalence of infection amongst younger aged groups (Sasidharan et al. 2011). This may be related to several risk factors documented in developing countries for early childhood exposure to *H. pylori,* such as the increased number of siblings, sharing of beds during childhood, poor hygiene, unclean water, and low socioeconomic status (Raymond et al. 2008). Gender was also not found to be significantly associated with *H. pylori* infection in our study. However, other local studies have reported a higher prevalence of *H. pylori* infection in males, compared to females (Alfizah et al. 2010; Chieng et al. 2015; Sasidharan et al. 2011). A recent meta-analysis of 244 studies had also reported that male sex was associated with a greater prevalence of *H. pylori* infection (Ibrahim et al. 2017).

Several local studies have consistently reported a much higher prevalence of *H. pylori* infection amongst Indians, followed by Chinese and Malays (Alfizah et al. 2010; Chieng et al. 2015; Goh 2009; Kaur & Naing 2003). Goh and Parasakhti (2001) proposed a racial cohort theory to explain the high prevalence rate amongst Indians and Chinese ethnicities in Malaysia. The high prevalence amongst Indian and Chinese ethnicities in Malaysia may be reflected by the high prevalence rates reported in Southern India (62%) and Southern China (44%). It is postulated, that these countries were the origin of these ethnicities, which had subsequently passed down the *H. pylor*i infection through generations born in Malaysia, or after their migration to Malaysia. This is further supported by the paucity of interracial marriages, leaving the ethnicities remaining fairly distinct, and encouraged transmission of infection within the racial group (Goh 2018; Pue & Sulaiman 2013). Other causes for the differences in prevalence amongst ethnicities may be attributed to the virulence factors of *H. pylori* strains, which are distinct to different ethnicities, host susceptibility factors, and

environmental factors, including diets peculiar to different ethnicities (Goh 2018). In our study, although we did not show any statistically significant difference between ethnicity and infection, the proportion of infected patients appeared to be highest among Indians.

The performance of our IRUT was comparable, and had a very good agreement with the CLO test, proving its status as an acceptable alternative to the CLO test for the diagnosis of the *H. pylori* infection. The slightly lower specificity of our IRUT may be attributed to the non-inclusion of the antibacterial agent, such as the one which was used in the CLO test. However, it is unlikely that sufficient amounts of urease-producing bacteria were present, or was thought to have overgrown in 20 min, to cause a false-positive result in the IRUT. Urease-producing gastric mucosal colonisers such as *Proteus mirabilis,* or *Klebsiella* species, commonly cause positive results if the tests were read at more than 24 h (Xia et al. 1994). False-positivity was also been reported with gastric biopsy specimens which were contaminated with alkaline bile, due to reflux resulting in a higher surface pH (Ng et al. 1997). Our buffered IRUT medium aimed a pH of 5.5, to prevent false-positive or false-negative results. In terms of the gastric biopsy sites, the biopsy specimens in our study were taken from only the corpus region, as per the current hospital standard practice. Patchy distribution of *H. pylori* in the stomach may cause sampling errors, leading to false-negatives. Although *H. pylori* colonisation typically predominates in the gastric antrum, some studies demonstrated that the colonisation in the corpus is as common as in the antrum, albeit with lower bacterial numbers (Kuipers et al. 1995). Siddique et al. (2008) demonstrated a much higher sensitivity of the rapid urease test, when increased numbers of antral samples were taken from one (52%) to four samples (96%). This also concurred with the work by Lee et al. (2013), which showed an increased sensitivity when two biopsy specimens were taken from the antrum and corpus regions (85.6%), in comparison to only a single antrum biopsy (64.6%), and a single corpus biopsy (69.5%).

The authors were aware that the extremely low numbers of *H. pylori*-positive cultures may contribute to a false high clarithromycin resistance rate in the study. *H. pylori* cultivation from gastric biopsy specimens is known to be technically challenging, and time consuming. It requires skilful handling to maintain the culture at the needed specific conditions. Other difficulties noted in *H. pylori* cultivation from gastric biopsy specimens reported in the literature includes *H. pylori* adhering tightly to epithelial cells, making their release difficult from tissues, which could result in false-negative results, characterised by poor or non-detectable growth (Peretz et al. 2015). Optimisation with trypsin-treated gastric biopsy has been shown to improve the diagnostic sensitivity of the *H. pylori* cultivation technique (Peretz et al. 2015).

Earlier Malaysian studies reported low clarithromycin resistance rates, ranging from 0 to 2.1% (Ahmad et al. 2011; Goh & Navaratnam 2011). A later study documented a much higher rate at 6.8% (Teh et al. 2014). In Asia, an increase in clarithromycin resistance was also reported from 15.28% to 32.46% over a 6-year period (Ghotaslou et al. 2015). It impacted the efficacy of clarithromycin-based triple therapies. Pooled resistance rates have in fact, been reported to have steadily increased over the years in the western pacific region (Savoldi et al. 2018). Clarithromycin resistance and eradication failures have been associated with previous exposure to macrolides (McMahon et al. 2003), and has been increasingly noted to drive point mutations in the 23S rRNA gene, that leads to a decrease in the macrolide binding (Occhialini et al. 1997). In this study, however, we did not elicit the history of previous clarithromycin usages, or non-compliance to previous eradication therapies amongst the patients. This should be researched in future studies, along with the genotypic determination of mutations responsible for clarithromycin resistance, such as the A2143G, A2142G and A2142C mutations (Alfizah et al. 2014).

Conclusion

The overall prevalence of *H. pylori* infection in Sungai Buloh Hospital, Malaysia, is low and coherent with other local studies which depicted similar ethnic compositions. However, the high proportion of clarithromycin-resistant isolates may not reflect the exact resistance rates due to the small number of positive cultures. No association was found between the age groups, gender and ethnicity amongst the patients with *H. pylori* infection. Our in-house rapid urease test is an acceptable alternative to the CLO test for the rapid diagnosis of *H. pylori* infection.

Acknowledgements

This study was funded by the UKMMC Fundamental Research Grant (FF-2015-060). The authors would like to thank the Director General of Health, Malaysia, for his permission to publish this article. We would also like to thank the Director of the Sungai Buloh Hospital, for allowing the study to be conducted in the endoscopy room, as well as the Microbiology Unit of Sungai Buloh Hospital.

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Received: 3 April 2019
Accepted: 23 September 2019