Changi General Hospital
SingHealth

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Changi General Hospital
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www.cgh.com.sg
Reg No 198904226R

Test Name	Helicobacter pylori PCR (+ prediction of clarithromycin resistance) [In-House Test]
Specimen Type	Tissue, gastric biopsy (in saline) Tissue, gastric biopsy (in CLO test cartridge)
Specimen Storage And Transport	**Tissue biopsy in saline:** **Transport to laboratory as quickly as possible.** Samples should be refrigerated, if delay in specimen transport is anticipated. Refrigerated samples may be stored and tested for up to approximately 1 day after specimen collection. **Tissue, gastric biopsy (in CLO test cartridge):** Transport to laboratory as quickly as possible. Samples may be stored and tested up to a maximum of 1 day after specimen collection.
Specimen Minimum Volume	-
Test Method	Real-time PCR (Lab developed test) (non-FDA approved)
Expected Result	Helicobacter pylori DNA detected; Helicobacter pylori DNA NOT detected. For samples where Helicobacter pylori DNA is reported as detected, the following additional results will be available: "Presumptive clarithromycin RESISTANT" "Presumptive clarithromycin RESISTANT – heterogenous population present" "Presumptive clarithromycin susceptible" "Clarithromycin resistance INDETERMINATE"
Reference Ranges	n/a
Turn Around Time	1-3 day
Days Of Testing	Monday - Friday, working hours
Hospital	CGH
Laboratory	Microbiology Lab
Discipline	Microbiology
Contact Details	6850 4917 / 6850 4935
CPOE Order Name Synonyms	H pylori PCR
Clinical Information	***Helicobacter pylori* detection** The PCR tests for the presence of a 23S rRNA sequence that is specific to *Helicobacter pylori*. *H. pylori* culture and isolation is time-consuming, requiring significant resources that makes this an expensive diagnostic method. However, bacterial culture derived from a gastric biopsy specimen is one of the most reliable methods, providing specificity up to 100%, although a lower sensitivity (around 90%) compared to that observed with histology and the rapid urea test. The high sensitivity of PCR-based methods can help detect *H. pylori* infection in patients with peptic ulcer bleeding, gastric cancer, or gastric MALT lymphoma for whom the diagnosis of *H. pylori* is important but difficult to obtain by other non-molecular methods. Previous studies have found that PCR-based methods can detect low-density infection in a number of patients with dyspepsia, compared with non-molecular conventional methods. False-positive PCR results may be observed as a consequence of residual nucleic acid following antimicrobial therapy, and PCR may not be reliable as a "test-of-cure". **Prediction of resistance to clarithromycin** Clarithromycin resistance in *H. pylori* is predominantly mediated by point mutations in domain V of the 23S rRNA gene, namely A2142G/C and A2143G mutations. The assay detects mutations present in a 26 base-pair region of the relevant domain V. If characteristic mutations are seen at A2142G/C or A2143G, the PCR will be reported as "Presumptive clarithromycin RESISTANT".

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	In our CGH-based study, 19% (11/58) of "clarithromycin-resistant" samples contained a heterogenous population of both genotypic clarithromycin-resistant and clarithromycin-susceptible *H. pylori* strains (i.e., a mixed population was present in the biopsy sample). These sample types will be reported as "Presumptive clarithromycin RESISTANT – heterogenous population present". If NO characteristic mutations are detected, the PCR will be reported as "Presumptive clarithromycin susceptible". In a small number of cases, atypical mutations in the tested 23SrRNA region may be present, where the impact on phenotypic clarithromycin resistance is unknown. If these atypical mutations are detected, the PCR will be reported as "Clarithromycin resistance INDETERMINATE". Point mutations in the domain V of 23S rRNA genetic region lead to bacterial resistance to macrolides. Common mutations implicated in clarithromycin resistance are A2143G, A2142G, and A2142C, in some studies confer resistance in 80–90%, 16–17%, and 2–4% of clinical isolates, respectively. These mutations are detected by this PCR. Several other mutations have uncommonly been reported in diverse geographical regions and are associated with the clarithromycin resistance phenotype. Because of the possibility of multiple mechanisms of clarithromycin, culture-based susceptibility testing (phenotypic) remains the gold standard. Genotypic (PCR-based) methods are more accurate than culture in detecting low numbers of resistant bacteria in a population of susceptible bacteria, which is termed "heteroresistance populations". Based on CGH data, 82% (n=9) of heteroresistant populations subsequently were culture positive for phenotypic clarithromycin-resistant H. pylori. A recent review identified that the prevalence of heteroresistance was 7%, while a genotypic based study reported that clarithromycin-containing therapy eradicated heteroresistant infections at a significantly lower rate in comparison with susceptible cases (P = .0112), but more effectively than homogenously resistant populations. Based on data from 261 isolates in CGH, the PCR-based method (genotypic) showed agreement with phenotypic susceptibility-based testing for 239 of isolates.
Link Out For Additional Information	
Remarks	**INFORMATION ON ASSAY PERFORMANCE CHARACTERISTICS** **Detection of *Helicobacter pylori*: sensitivity and specificity** A total of 410 gastric tissue biopsies were cultured for *H. pylori* and concurrently tested by the PCR assay. Among these samples, 407 samples had a valid PCR result. The 270 culture-positive samples were also positive for *H. pylori* by PCR, while 113 samples were *H. pylori*-negative by both PCR and culture. Twenty-four samples were *H. pylori*-positive by PCR but negative by culture. There were no tissue samples that were H. pylori-positive by culture but negative by PCR. 16S rRNA sequencing was performed from the first three *H. pylori* PCR positive / culture-negative samples; the 16S rRNA sequences derived from all three samples were consistent with *H. pylori*. **Prediction of resistance to clarithromycin** Phenotypic clarithromycin susceptibility results were available for 261 samples. In the PCR group which were reported as "Presumptive clarithromycin susceptible" (n=191), 183 (96%) samples were concordant with phenotypic result (clarithromycin susceptible by conventional susceptibility testing). For the PCR group reported as "Presumptive clarithromycin RESISTANCE" (n=58), 56 (97%) samples were concordant with phenotypic result (clarithromycin resistant by conventional susceptibility testing). **LIMITATIONS OF ASSAY** The performance of the test may be affected by inappropriate sample collection, handling and transportation. The effect of antibiotic treatment on test performance has not been evaluated. Bacterial nucleic acids may persist in vivo independent of organism viability. Detection of organism target(s) does not imply that the corresponding organisms are infectious.

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