The 2005 Nobel Prize in Physiology or Medicine: Helicobacter pylori and its role in gastritis and peptic ulcer disease

Gastritis (inflammation of the stomach) and peptic ulcer disease (PUD; ulceration of the stomach or duodenum) have been a major human health problem the world over, particularly in most countries of the developed world. In USA and Australia, one in ten people might be expected to suffer from PUD during one’s lifetime; and in the developing world, the incidence of the disease seems to be still higher, although similar data are not available. In the past, it was believed that gastritis and peptic ulcers were caused by acid, stress, spicy food, etc. and should be treated by drugs blocking acid production. However, these traditional antacids ulcer medications provided only temporary relief. This situation dramatically changed, when it was discovered that gastritis and PUD are actually caused by a bacterium (later named Helicobacter pylori), although some ulcers are also caused by long-term use of non-steroidal anti-inflammatory agents (NSAIDs) like aspirin and ibuprofen. When caused due to bacteria, the patients suffering with this disease could be permanently cured by eradication of H. pylori, using antibacterial therapy\(^1\). There is evidence that H. pylori infection also causes gastric cancer, the adenocarcinoma, which can be prevented by H. pylori eradication, although convincing evidence based on clinical trials is lacking.

It is also known now that H. pylori is a ubiquitous gastrointestinal organism, which infects over three billion people the world over\(^2\), with the infection generally becoming less common with rise in the standard of living\(^3\) (Figure 1). A surprising finding, however, was that not more than 20% of people (regardless of age) who tested positive for H. pylori had ulcers.

The above pioneering work involving discovery of H. pylori and its role in the development of gastritis and PUD has been recognized by the award of 2005 Nobel Prize for Physiology or Medicine to two Australian scientists, Barry J. Marshall working at the Queen Elizabeth II Medical Center of the University of Western Australia (Perth), and J. Robin Warren, who retired in 1999 from his position as a senior pathologist at the Royal Perth Hospital, also located in Perth, Western Australia.

**Life of Barry Marshall**

Barry Marshall was born on 30 September 1951 in Kalgoorlie, Western Australia, as the eldest of four children in a family with no privileged background (his father was a boiler-maker, and his mother was a nurse). Marshall studied medicine at the University of Western Australia during 1968–74 to earn the MBBS degree. Later he worked in Royal Perth Hospital, during 1977–84 as Registrar, Medicine, and during 1985–86 as NHMRC Research Fellow, Gastroentrology. In 1986, Marshall moved out to USA and worked at the University of Virginia, first as a Research Fellow and Professor of Medicine (1986–94), and later as Professor of Research in Internal Medicine (1996). In 1996, Marshall returned from USA to Perth as an internationally acclaimed medical scientist and then worked at his alma mater, the University of Western Australia in various capacities.

**Awards and recognitions won by Warren and Marshall**

Warren and Marshall jointly won the following awards: Warren Alpert Prize of Harvard Medical School (1995), Australian Medical Association Award (1993), Paul Ehrlich and Ludwig Darmstaedter German Medical Research Prize (1997), and the inaugural Florey Medal Award (1998). Both researchers were also honoured individually. Warren received the Distinguished Fellows Award of the College of Pathologists (1995), Australian Medical Association (WA) Award (1995), Medal of the University of Hiroshima (1996), Award of the First Western Pacific Helicobacter Congress (1996), and honorary degree of Doctor of Medicine from the University of Western Australia (1997). Marshall won Albert Lasker Award (1995), Australian Achiever Award (1998), Burt Fellowship of the National Health and Medical Research Council (1998),

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**Figure 1.** Map showing percentages of population infected with Helicobacter pylori. Rates of infection vary in different regions, and can be attributed to socio-economic conditions in childhood. As populations achieve better living conditions, incidence of H. pylori decreases (from the Helicobacter Foundation website: www.helico.com).
Table 1. Summary of work by Warren and Marshall on *H. pylori* in chronological order

<table>
<thead>
<tr>
<th>Event</th>
<th>Description</th>
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<tr>
<td>Noticed that inflammation of the stomach (gastritis) was associated with the presence of a bacterium (Warren).</td>
<td>Studied 100 patients and discovered that this bacterium was present in every patient who suffered from duodenal ulcer (Warren and Marshall).</td>
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<td>Grew the first culture of the bacterium, which was later named <em>H. pylori</em> (Marshall and microbiologists from Royal Perth Hospital).</td>
<td>Swallowed a culture of this bacterium, and suffered acute symptoms in order to prove the hypothesis that <em>H. pylori</em> was the cause of gastritis (Marshall and a volunteer).</td>
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<td>Promoted this hypothesis, despite significant scepticism from gastroenterology specialists (Marshall).</td>
<td>Through persistence and publication of research papers, stimulated much research and treatment trials which eventually proved that <em>H. pylori</em> did indeed cause gastritis and gastric ulcers (Warren and Marshall).</td>
</tr>
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**Initial discovery of Helicobacter pylori and fruitful partnership between Warren and Marshall**

In 1979, for the first time, Warren observed the presence of small curved bacteria on a biopsy of the gastric mucosa. Later in 1981, Warren met Marshall, and a fruitful partnership followed, which demonstrated the clinical significance of the bacteria\(^6\).\(^\text{-}^9\) (see later for details).

**Initial scepticism about the discovery**

In the early 1980s, Warren and Marshall were the subject of criticism and unfounded jokes, since everyone was taught and believed that bacteria could not survive in the stomach’s acid environment. But Barry was aggressive in selling his message, although he had to struggle to get his work presented and published, and it took them at least a decade longer than expected to convince the scientific world that peptic ulcer was caused by *H. pylori*.

**Koch’s postulates fulfilled to establish link between gastritis/ PUD and *H. pylori***

Warren and Marshall’s work almost fulfilled all four postulates of Koch\(^10\) (but see below). A summary of the work done by Warren and Marshall to fulfill Koch’s postulates is presented in Table 1.

**Association of *H. pylori* with disease**

Warren and Marshall commenced their research by studying a group of 100 patients who had undergone endoscopy for gastric conditions\(^6\).\(^\text{-}^8\). They reconfirmed the link between gastritis and presence of the bacterium, thus satisfying the Koch’s first postulate.

**Culturing of *H. pylori* as a new species**

For culturing the bug, Warren and Marshall initially used without success, the selective growth conditions appropriate for *Campylobacter*. Later, in 1982, when the cultures were inadvertently left in the incubator over the Easter holidays (due to lack of laboratory staff), chance prolongation of the incubation period from the usual 2 to 6 days resulted in successful growth and isolation of the bacterium\(^6\). This led to the discovery that the growth of *H. pylori* was much slower than other bacteria and allowed culturing of the bug, thus satisfying Koch’s second postulate.

The bug causing gastritis and PUD was initially named *Campylobacter pyloridis* due to its morphological similarity with members of the genus *Campylobacter*, and due to its isolation from the pyloric region of the stomach\(^6\).\(^\text{-}^9\). The name of the bacterium was grammatically corrected in 1987 to *C. pylori* and, in 1989, it was re-named *Helicobacter pylori* (Figure 2), which became the type species of the new genus *Helicobacter*, which is now known to have several species that infect animals other than humans\(^11\). These species can be distinguished with the help of 16S rRNA sequence.

**Marshall swallowed a culture of *H. pylori* to prove his hypothesis**

In order to satisfy the third and fourth postulates of Koch, Marshall himself and a volunteer swallowed a solution containing the bug to prove that it caused the disease. About a week later, Marshall started vomiting and showing other painful symptoms of gastritis. The biopsies revealed that he had developed both gastritis and an infection with *H. pylori* (Figure 3), but failed to develop an ulcer, and the disease resolved without treatment\(^12\),\(^13\).

Consequently, although this constituted highly suggestive evidence that the organism caused gastritis, it was far from conclusive, so that as late as 1995, Marshall himself conceded that all Koch’s postulates had not been fulfilled for confirmation of a causal relationship between *H. pylori* and PUD\(^14\). A more important confirmation, however, was the eradication of *H. pylori* through the use of antibacterial drugs.

**Association of *H. pylori* with gastric cancer and cardiovascular diseases?**

Even before the discovery of *H. pylori*, an association between cancer of the stomach and chronic gastritis was observed, which suggested a possible link between *Helicobacter* infection and cancer\(^15\),\(^\text{-}^17\). Similarly, several studies have also suggested an association between *H. pylori* infection and an increased risk of cardiovascular disease\(^18\),\(^\text{-}^21\), although no causal relationship could be established\(^22\).

**How could the bug grow in acidic environment of the stomach?**

The question remained as to how *H. pylori* could survive in the acidic environment.
of the stomach. Warren showed that the bacteria grow on the surface epithelium, covered with a thick layer of normal mucus. In addition, Marshall found that the bacteria produce a large amount of urease, an enzyme that breaks down urea into ammonia and carbon dioxide to form a protective alkaline layer around them.

Diagnosis of H. pylori-related ulcers

Diagnosis of ulcers

Once symptoms of an ulcer (e.g. abdominal discomfort, weight loss, poor appetite, nausea, etc.) are observed, the doctor may conduct an upper gastrointestinal (GI) series (including X-ray of the oesophagus, stomach and duodenum) or an endoscopy/biopsy. Once GI and/or endoscopy confirms the presence of ulcer, the patient is tested for H. pylori, since ulcers may also be caused by NSAIDs.

Diagnosis of H. pylori

The bug H. pylori may be diagnosed through blood, breath, stool and tissue tests. The urease test allows patients to have a diagnosis within 20 minutes of having a biopsy, and start curative therapy immediately. In non-invasive breath test, the patient is made to swallow a small amount of urea labelled with a carbon isotope ($^{13}$C or $^{14}$C), which is broken down by H. pylori to release carbon dioxide in the breath; this breath test became a popular and accurate means of diagnosing H. pylori in patients.

Treatment for H. pylori eradication?

It was shown that 80% of patients were permanently cured of their ulcer, if H. pylori was eradicated. This resulted in a complete reassessment of ulcer treatment, and became an essential part of the management of ulcer disease. Therapy for H. pylori infection consists of 10 days to 2 weeks of one or two effective antibiotics, plus either a H$_2$ blocker or a proton pump inhibitor; a stomach lining protector may also be used (Table 2). Currently, eight H. pylori treatment regimens are approved (Table 3), and the eradication rates range from 61 to 94%. Overall, triple therapy regimens have shown better eradication rates than dual or quadruple therapy. Longer time of treatment (14 versus 10 days) results in better eradication rates.

Dual, triple and quadruple therapy

Two weeks of dual therapy involving two drugs (an antibiotic and an acid suppressor) is generally recommended for eradication of H. pylori. However, it is not as effective as triple therapy, which involves taking two antibiotics to kill the bacteria and either an acid suppressor or stomach-lining shield. Two weeks of quadruple therapy (also called bismuth-triple therapy), which uses two antibiotics, an acid suppressor and a stomach-lining shield, was also found promising in several studies. Despite some problems and side effects associated with triple therapy, recent studies show that it is ideal.

Whole genome sequence and genetic variability of H. pylori

In 1997, the genome (17 megabase) of H. pylori was fully sequenced, and found to have 1600 genes, of which only 750 are essential. At least 23% of all genes are unique to H. pylori, which should be specific for survival of the pathogen in the stomach. A high degree of genetic variability also exists among different strains of H. pylori, which explains why only one in six infected humans suffer from ulcers. About 60% of H. pylori strains in USA contained a particular sequence of genes, i.e. a 'pathogenicity island' (encoding Cag-PAI), and people infected with a strain containing this pathogenicity island were significantly more likely to develop ulcers or adenocarcinoma than those infec-
Table 3. FDA-approved treatment options

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<tr>
<td>Omeprazole 40 mg OD + clarithromycin 500 mg TID x 2 wks, then omeprazole 20 mg OD x 2 wks.</td>
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<tr>
<td>Ranitidine bismuth citrate (RBC) 400 mg BID + clarithromycin 500 mg TID x 2 wks, then RBC 400 mg BID x 2 wks.</td>
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<tr>
<td>Bismuth subsalicylate (Pepto Bismol®) 525 mg QID + metronidazole 250 mg QID + tetracycline 500 mg QID x 2 wks + H₂ receptor antagonist therapy as directed x 4 wks.</td>
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<tr>
<td>Lansoprazole 30 mg BID + amoxicillin 1 g BID + clarithromycin 500 mg TID x 10 days.</td>
</tr>
<tr>
<td>Lansoprazole 30 mg TID + amoxicillin 1 g TID x 2 wks.</td>
</tr>
<tr>
<td>Lansoprazole 50 mg BID + clarithromycin 500 mg BID x 2 wks.</td>
</tr>
<tr>
<td>Lansoprazole 50 mg BID + clarithromycin 500 mg BID + amoxicillin 1 g BID x 10 days.</td>
</tr>
<tr>
<td>Lansoprazole 50 mg BID + clarithromycin 500 mg BID + amoxicillin 1 g BID x 10 days.</td>
</tr>
</tbody>
</table>

*Although not FDA-approved, amoxicillin has been substituted for tetracycline in patients for whom tetracycline is not recommended.

**This dual therapy regimen has restrictive labelling. It is recommended for patients who are either allergic or intolerant to clarithromycin or for infections with known or suspected resistance to clarithromycin.

The cytoxin-associated genes A and E (caga, cageE), and the vacuolating cytotoxin gene A (vacA) of H. pylori have been associated with phenotype characteristics of virulence. Therefore, CagA, CagE, Cag-PAI and VacA are also considered potential targets of vaccine development against H. pylori.

The H. pylori Foundation: A source for additional information

The ‘Helicobacter Foundation’ was founded by Barry Marshall in early 1994. He chartered the Foundation in order to provide people with information on H. pylori and its effects. The Helicobacter Site (www.helicob.com) is maintained by Marshall and ConcreteBob Software. An upgrade is currently under way. Readers may get additional information on this site.


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