Two Tall Poppies and the Discovery of *Helicobacter Pylori*

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J Robin Warren, the microscopist, June 11, 1979

Pathologist J Robin Warren at Royal Perth Hospital in Western Australia, on examining a gastric biopsy specimen, noticed a blue line on the surface of the gastric mucosa. Higher magnification of the fresh tissue revealed numerous small bacilli closely adherent to the epithelium. Such well fixed gastric biopsies were rare before endoscopy became commonplace in the 1970s. Gastric resections were placed unopened in formalin fixative, and slides of the mucosa showed moderate autolysis and little detail. Postmortem specimens rarely revealed potential bacteria, and any fine detail of the gastric mucosa was rarely preserved due to enzyme and acid digestion. The appearance of the fiberoptic endoscope in the 1970s allowed for retrieval and fixation of well-preserved gastric mucosal specimens. Warren was applying various bacterial stains to these specimens. His first attempt with the Warthin-Starry silver stain revealed numerous bacilli that were clearly visible, even on low power magnification[1] (Fig. 1).

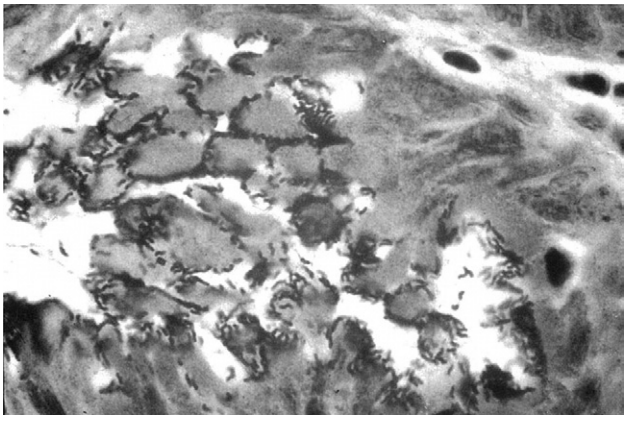

Figure 1. The first time ever Dr Warren visualized *Helicobacter pylori*.

Barry J Marshall, the clinical collaborator, July 1981

An internal medicine trainee, Barry Marshall was assigned to gastroenterology and asked the division head, Dr Tom Waters, about doing a research project. Waters presented him with a list of 25 patient names that had been given to him by one of the pathologists, Dr Robin Warren, who had been looking for a clinical collaborator interested in following up on some bacteria he had seen on stomach biopsies. Marshall reviewed the list and was intrigued to see a patient whom he had referred for endoscopy during a previous internal medicine rotation, diagnosed with "nodular gastritis of the antrum." He met with Warren and the two spent an afternoon reviewing slides of the curved bacteria easily seen using the Warthin-Starry silver stain, and they discussed the histopathology of the gastric mucosa. Marshall was aware that the stomach was thought to be sterile, but Warren presented such a convincing case implicating the bacteria as a pathogen that Marshall accepted the concept, hypothesized that this was a new species of bacteria, and surmised that the discovery would make for a "nice publication, regardless of whether or not it actually caused a disease." Marshall was familiar with the published literature describing *Campylobacter jejuni* as a cause of foodborne gastroenteritis, and both he and Warren appreciated that these newly discovered gastric organisms had a similar shape. Marshall reviewed the records of Warren's 25 patients and coded the clinical pathologic findings that could be related to the spiral bacteria. All 25 patients had campylobacter-like organisms (CLO) on biopsy and the endoscopic diagnoses were gastritis (n = 12), gastric ulcer (n = 7), erosions (n = 4), and duodenal ulcer (n = 2).[2]

Serendipitous CLO culture, April 1982

Standard attempts to culture the organisms were unsuccessful. In order to obtain fresh specimens, Marshall "hand-delivered" gastric biopsy specimens from 20 patients suspected as having gastritis directly to the microbiology laboratory. As with many important discoveries, the first successful culture of the CLO occurred serendipitously. The microbiology section that performed feces and throat swab cultures also handled gastric biopsy specimens. Standard practice was to discard the plates if no growth was seen within 48 hours. Due to the 4-day Easter holiday in 1982, a gastric biopsy remained in the incubator from April 8 through April 13, allowing for a 5-day incubation period. When the technician finally examined the plate, small transparent colonies of CLO were readily evident. Protocol

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Abbreviations and Acronyms

CBS = colloidal bismuth subcitrate
CLO = campylobacter-like organisms
CP = *Campylobacter pylori*
HP = *Helicobacter pylori*

changed to incubate all gastric biopsy plates for 4 days. In subsequent weeks, more of the CLOs were isolated from patients.[2]

The Gastroenterologic Society of Australia abstract rejection, February 1983

Marshall and Warren submitted an abstract to the Gastroenterologic Society of Australia for the upcoming May meeting in Perth. The abstract was a clinical trial of 100 patients who underwent elective gastroscopy with antral biopsies. Spiral bacteria were present in the mucosa of patients with active chronic gastritis, duodenal ulcers, and gastric ulcers. The authors related that microbiologic and electron microscopic evidence supported the thesis that these bacteria were a new genus and etiologically associated with gastritis and gastritis-associated diseases. Their abstract was rejected, and that March, the condolence letter from the secretary stated that "a large number of high quality abstracts made it extremely difficult" to select those for presentation.[2]

David McGechie, a microbiologist and colleague, suggested that the microbiology community may be more interested in the organism and recommended that Marshall contact Martin Skirrow in England. Skirrow requested that Marshall send him some cultures and invited him to submit an abstract for presentation at the Second International Workshop on Campylobacter Infections in Brussels that September.[2] Marshall attended the workshop and his presentation aroused a great deal of interest in the flagellated spiral bacteria of the stomach.[3]

Lancet, June 1983

Lancet published Warren and Marshall's observations with the bacteria in the form of 2 letters. Warren commented that in patients with active chronic gastritis, the curved bacilli were nearly always present in large numbers, growing between the cells of the surface epithelium. Marshall related that they could routinely culture the bacteria and the morphologic characteristics, "up to five sheathed flagella arising from one end" indicated that they were a new species. He stated that the pathogenicity of these bacteria remained unproved, but if these bacteria proved to be associated with gastritis, they could also be associated with peptic ulcer disease and gastric cancer.[4]

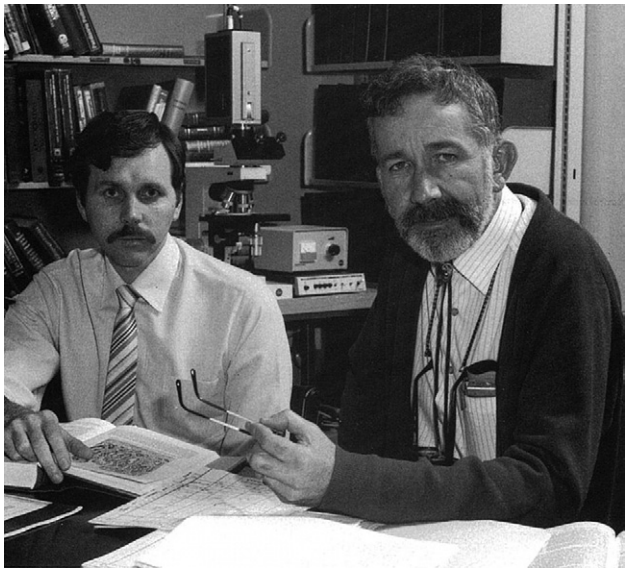

Figure 2. Drs Barry J Marshall (left) and J Robin Warren, July 1984.

A changing tide, January 1983 to September 1984

Marshall, McGechie, Rogers, and Glancy isolated the organism from 114 patient antral biopsies. All isolates were sensitive to a variety of commonly used antibiotics and bismuth. Eighty percent were sensitive to metronidazole or tinidazole. Marshall suggested that antibacterial regimens could provide a cure for dyspeptic disease and peptic ulceration.[5]

Lancet published Marshall and Warren's study of 100 consecutive gastroscopic biopsy specimens, 58 of which demonstrated the curved bacilli. Thirty-one specimens showed chronic gastritis and 27 had gastric or duodenal ulcers on endoscopy. They concluded that "pyloric campylobacter is aetiologically related to chronic antral gastritis and, probably, to peptic ulceration"[6] (Fig. 2).

A skeptical scientific world, January to June 1984

Prompted by the 1983 letters in *Lancet* and Marshall's presentation at the Second International Workshop on Campylobacter Infections in Brussels, 4 publications appeared discounting the findings of Marshall and Warren. McNulty and Watson[3] concluded that there was a "strong correlation between bacterial colonization and gastritis and peptic ulcer; however, their pathogenic role, if any, is obscure." Eldrige and colleagues[7] stated that "the frequent association of this organism with gastritis cannot be taken as evidence of any pathologic role for CLO." Likewise Langenberg and associates[8] opined that "CLO-associated gastritis can be present in apparently healthy individuals, which seems to put into question its clinical significance." Finally, Rollason and coworkers[9] firmly stated that it is