Microbiological, Biochemical and Functional Characterization of Helicobacter pylori infection.

Review article

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INTRODUCTION

In 1979 Robin Warren, a pathologist in Perth, Western Australia, began to notice that curved bacteria were often present in gastric biopsy specimens submitted for histological examination. These organisms were not present within the gastric mucosa but were present in the mucus layer overlying the tissue [2]. Warren found that because they could never be isolated, they were ignored and ultimately forgotten by generations of physicians and scientists. A young trainee in internal medicine, Barry Marshall, became interested in Warren’s observations, and together the two sought to isolate the organisms from biopsy specimens. Since the organisms had the appearance of curved, gram-negative rods, the investigators used methods for the isolation of Campylobacter species, including inoculating the biopsy specimens on to selective media and incubating the cultures under microaerobic conditions. Since most Campylobacters grow within 48 h under such conditions, plates without visible growth were discarded within 3 days. The initial cultures from approximately 30 patients were negative, but by chance one culture was incubated for 5 days over an Easter holiday and colonies were seen [60] subsequently, organisms were isolated from 11 patients. The organism was characterized and called Campylobacter pyloridis (now known as Helicobacter pylori). Following publication of this seminal report, investigators all over the world rapidly confirmed the presence of these organisms in the gastric mucus [62]. It had become clear that H. pylori infection was strongly associated with the presence of inflammation in the gastric mucosa (chronic superficial gastritis), and especially with polymorphonuclear cell infiltration (chronic active gastritis), [31].

Marshall and Warren noted that H. pylori infection was associated with duodenal ulceration [60], and this observation too was rapidly confirmed and extended to include gastric ulceration [30]. By 1994, the National Institutes of Health conference concluded that H. pylori was a major cause of peptic ulcer disease and recommended that infected individuals with ulcers should be treated to eradicate the organism [74]. Several evidences had indicated that chronic gastritis is linked to the development of adeno carcinoma of the stomach, the most important gastric malignancy in the world [57], but the causation of the gastritis was then unknown. In 1991, four reports first showed associations between H. pylori infection and the presence or the development of gastric cancer [86,123]. In 1994, the International Agency for Cancer Research, of the World Health Organization, reviewed the available evidence and declared that H. pylori was a carcinogen of humans [26].
Microbiological characteristics of *H. pylori*:

Morphology: *H. pylori* is a gram-negative bacterium with bluntly rounded ends in gastric biopsy specimens, measuring 2 to 4 μm in length and 0.5 to 1 μm in width. Although usually spiral-shaped, the bacterium can appear as a rod [49]. However, when cultured on solid medium, the bacteria assume a rod-like shape, spiral shapes are infrequent or absent [48]. After prolonged culture on solid or in liquid medium, coccoid forms typically predominate [38]. By electron microscopy, coccoid forms appear as U-shaped bacilli with the ends of the two arms joined by a membranous structure. Coccoid forms are metabolically active; however, they cannot be cultured in vitro [38]. In gastric biopsy specimens, *H. pylori* organisms are 2.5 to 5.0 mm long and 0.5 to 1.0 mm wide; there are four to six unipolar sheathed flagella [48], which are essential for bacterial motility. Flagella exhibit a characteristic terminal bulb, which is an extension of the flagellar sheath [128]. The flagellar sheath exhibits the typical bilayer structure of a membrane [128]. Ultrastructurally, when tannic acid is used as a mordant, it can be seen that the outer membrane of *H. pylori* is coated with a glycolocalyx-like structure; in gastric biopsy specimens, the surface of individual bacteria may be linked to gastric epithelial microvilli by thread-like extensions of the glycolocalyx [49]. The surface of viable *H. pylori* cells grown on agar plates is coated with 12- to 15-nm ring-shaped aggregates of urease and HspB, a homolog of the GroEL heat shock protein [28]. Urease and HspB are also associated with the surface of viable *H. pylori* invivo [43].

Respiration and Metabolism:

In tests involving routine microbiological methods, *H. pylori* does not appear to utilize carbohydrates either fermentatively or oxidatively [48]. However, studies begun to elucidate metabolic pathways in *H. pylori*. The studied isolates exhibit glucose kinase activity [65], which is associated with the bacterial cell membrane. In addition, enzyme activity characteristic of the pentose phosphate pathway has been identified [65]. Thus, *H. pylori* appears to be capable of catabolizing D-glucose. *H. pylori* possess specific D-glucose transporters; some characteristics of the glucose transport system appear to be unique [64].

*H. pylori* exhibit urea cycle activity, which may serve as an effective mechanism to extrude excess nitrogen from bacterial cells [66]. The Entner-Doudoroff pathway has been demonstrated in *H. pylori* [67]. Fumarate reductase is an essential component of the metabolism of *H. pylori* and as such constitutes a possible target for therapeutic intervention [68]. *H. pylori* can metabolize amino acids by fermentative pathways similar to those in anaerobic bacteria [221]. Cytochromes involved in termination of the respiratory chain of *H. pylori* have been characterized [59, 73]. The elevated level of CO2 required for growth of *H. pylori* in vitro may be due in part to activity of the enzyme acetyl coenzyme A carboxylase [34].

*H. pylori* cells contain polyphosphate granules, which may function as a reserve energy source in bacteria associated with a degenerated epithelium, where an exogenous energy source may be absent [32, 36]. Such studies of the metabolism of *H. pylori* are rapidly expanding our basic understanding of this important gastric pathogen; a practical outcome may be the identification of unique metabolic pathways which may serve as therapeutic targets with minimal effect upon the host [33].

Metabolism: *H. pylori* can be grown only in chemically defined medium with the additional amino acids arginine, histidine, isoleucine, leucine, methionine, phenylalanine and valine, and some strains also require alanine and/or serine [21]. *H. pylori* are urease, catalase, and oxidase positive, characteristics which are often used in
identification of *H. pylori*. *H. pylori* can catabolize glucose, and both genomic and biochemical information indicates that other sugars cannot be catabolized by *H. pylori* [5, 12].

(i) Oxidative stress defense: *H. pylori* is a micro aerophilic bacterium that does not tolerate high oxygen conditions, but it requires at least 2% O₂. This is because *H. pylori* uses oxygen as a terminal electron acceptor. *H. pylori* cannot utilize alternative electron acceptors, such as nitrate or formate [15].

(ii) Nitrogen metabolism: The gastric environment contains amino acids and urea as two major sources of nitrogen. Ammonia is a key component in nitrogen metabolism as well as acid resistance [22]. Consequently, *H. pylori* can utilize several alternative sources of ammonia [16, 24]. Also, *H. pylori* produces large amounts of urease, which has been estimated as up to 10% of the total protein content of *H. pylori*. Urease is a nickel-containing enzyme that consists of 12 UreA and 12 UreB subunits [10]. The UreA and UreB subunits have molecular masses of 27 kDa and 62 kDa [6].

(iii) Metal metabolism: Metals which are involved in a wide variety of chemical reactions. Metals are cofactors of enzymes, catalyzing basic functions such as electron transport, redox reactions, and energy metabolism, and are essential for maintaining the osmotic pressure of the cell. Consequently, they play an important role in the metabolism of all organisms. Since both metal limitation and metal overload delay growth and can cause cell death, metal homeostasis is of critical importance to all living organisms.

(a) Nickel: *H. pylori* requires efficient acquisition of nickel, as it is the metal cofactor of the essential colonization factors urease and hydrogenase. Nickel availability in human serum is very low (2 to 11 nM), and the nickel concentration in ingested food varies significantly depending on the diet and on food sources [3].

(b) Iron: In human or animal host tissues, the concentration of free iron is too low to support bacterial growth, as most iron is complexed into hemoglobin or chelated by transferrin in serum or by lactoferrin at mucosal surfaces. Iron sources available in the gastric mucosa are lactoferrin, heme compounds released from damaged tissues, and iron derived from pepsin-degraded food. It is thought that metals such as iron display increased solubility under the acidic conditions of the gastric mucosa and that eukaryotic iron-complexing proteins display lowered binding affinity under these conditions. The *H. pylori* genome encodes 11 proteins predicted to be involved in iron transport and 2 proteins thought to function as iron storage proteins [23].

(c) Copper: Copper is a cofactor for several proteins involved in electron transport, oxidases, and hydroxylases, but may also contribute to the formation of reactive oxygen species. *H. pylori* expresses several proteins which either are involved in copper transport or may act as copper chaperones [20].

(d) Cobalt: Cobalt is a trace metal cofactor of the arginase enzyme, which plays an important role in nitrogen metabolism of *H. pylori* [13] but also in modulating the immune response to *H. pylori* [8,9]. It has been suggested that *H. pylori* is exquisitely sensitive to cobalt in vitro [4], and it has been mentioned that cobalt may be used in non antibiotic therapy of *H. pylori* infections [1].

Biochemical and functional characterization:

Host inflammatory responses and effector molecules produced by *H. pylori* infected cells cause changes in the gastric mucosa during infection, requiring *H. pylori* to be able to adapt to an ever-changing environment. Several factors with vital roles in the colonization of *H. pylori* and the development of disease have been described.
These factors include the urease enzyme, flagella, the vacuolating cytotoxin VacA and the cag-pathogenicity island (cagPAI), which encodes the CagA protein and a type IV secretion (T4S) machinery that translocates CagA into host cells [151; 139-142]. Both phosphorylated and non phosphorylated CagA have been shown to interfere with host signalling pathways and cellular functions [110 108; 155]. Adherence is important for persistent infection of a host and is mediated by the binding of bacteria to glycoproteins and/or glycolipids present on the host cell surface. Analysis of gastric biopsy material has shown that H. pylori populations are found deep in the mucous layer, and a subset is attached to the surface of epithelial cells [109]. Recent reports have also suggested H. pylori to be localized intracellularly [152; 143 144; 90]. Several receptor structures for adherence of H. pylori to human gastric epithelial cells have been described, suggesting that H.pylori exhibits a multitude of adherence modes, which may change as infection progresses [106]. Two functional receptors for H. pylori, fucosylated ABO/Lewis bantigens (Leb) [100, 90] and sialyl-Lewis x/a antigens (sLex/a) human gastric epithelium, only minute levels of sialylated glycans are detected, but upon infection by H. pylori, increased expression of the inflammation/selectin as associated sLex occurs [133, 121]. Bacterial adherence to host cells is mediated by adhesins on the bacterial cell surface. Correspondingly, the H. pylori blood group antigen binding adhesin (BabA) mediates binding to the ABO/Leb receptor structures [114], and binding to sLex/a is mediated by the sialic acid binding adhesin (SabA) [133, 90].

Gram-negative bacteria continuously shed vesicles from the cell surface during their growth. These vesicles are 20–300 nm in size and are surrounded by an outer membrane (OM) layer, which is described to have a composition derived from the bacteria’s OM. Upon shedding from the bacterial cell surface, some of the under lying periplasmic and cytoplasmic proteins, as well as DNA and RNA, are contained within the vesicles [98,124]. The biological role of these vesicles has not been fully elucidated, but is described to be involved in toxin delivery, protein and DNA transfer, and signalling between bacteria. Biochemical characterization of vesicles isolated from pathogenic species has identified virulence associated factors such as toxins, invasins and host–effector molecules [112]. Electron micrographs of plasma from a person infected with Neisseria meningitides sero group B that died of septic shock showed that numerous vesicles were present [136]. H. pylori vesicles were found in gastric mucosal biopsy material and shown to contain the Vac Acyto toxin [104]. The protein content of H. pylori vesicles differs significantly from whole bacterial cells as well as of the OM. Using immunoblots, the presence of aseries of virulence factors in the vesicles such as adherence components and the oncoprotein CagA was confirmed [98].

The biochemical characterization and functional analysis of adherence components present on the surface of H. pylori vesicles was studied. Receptor conjugates for electron microscopy studies and an in vitro assay to characterize adherence to human gastric tissue sections was used. The results of these studies provide valuable insight into the mechanisms involved in bacterial–host interactions as they relate to H. pylori vesicles and their intimate interaction with human gastric mucosa.

Biochemical characterization of H. pylori vesicles:

(i) Characterization of major phospholipid vesicle components:

Phospholipids present in H. pylori vesicles were identified using two-dimensional (2D) 31P,1H NMR correlation spectra. The type and abundance of phospholipid components present in isolated vesicles, outer and inner membrane (IM) fractions, and whole H. pylori cells was determined. By definition, IMs have fewer proteins than OMs, thereby allowing IMs to be separated from OMs using density gradient centrifugation. Lipids were extracted
from whole bacteria, OM, IM and purified vesicles for fractions localization. For analysis of the 2D spectra, 1D projections were integrated. Phosphatidyl ethanolamine (PE) and cardiolipin (CL) were identified as the major phospholipids present in all fractions. IM fractions contained considerably lower amounts of phosphatidyl glycerol (PG) and lyso-phosphatidyl etanolamine (LPE) compared with the OM or vesicle fractions. Phosphatidyl choline (PC) was below the detection limit in IM, but was readily detected in all other fractions. These data represent the differences in phospholipid composition between the analysed fractions, and suggests that the phospholipid composition of vesicles most closely resembles that of the OM. Helicobacter pylori membranes have been reported to contain cholesterol [115]. The molar fraction of cholesterol estimated to be 10% relative to that of total phospholipids present in both whole bacterial cells as well as in vesicles.

(ii) Characterization of major proteins present in vesicles:

The major protein components of H. pylori vesicles were analysed using mass spectrometry (MS). Since many of the H. pylori OM proteins are of similar molecular masses, exhibit high pI values and do not resolve on 2D gels, 1DSDS-PAGE was combined with analysis by the sensitive nanoflow LC FT-ICR MS/MS method. Using H. pylori vesicles purified according to density, samples with adensity of 1.15–1.20 g ml-1 from fractions 3–9 were pooled. Initial separation of the samples by SDS-PAGE identified a series of 34 bands that were excised and subjected to peptide tandem mass determination. All mass data obtained were searched by MASCOT against all species in the NCBI database. An H. pylori proteome consists of approximately 1500 putative proteins [89, 143, 97], and in total 306 different H. pylori proteins were identified in the 34 excised bands. Identified vesicle proteins were classified according to clusters of orthologous groups (COG) for the total H. pylori proteome. Several H. pylori OM proteins have been associated with gastric disease [162], and therefore H. pylori proteins are listed as a separate group as classified according to [89]. A majority of all OM proteins (77%) were identified in the vesicles. Several of the identified vesicle proteins are associated with important roles in H. pylori colonization and virulence, such as urease subunits, the cytotoxin VacA, CagA, and the BabA and SabA adhesins. Also, g-glutamyl transpeptidase has been shown to exhibit immunosuppressive effects similar to the VacA protein [150] and the protease HtrA [130]. Among the series of cytoplasmic proteins GroEL, catalase, metabolic and ribosomal proteins were found. A second MS analysis (MS II) was performed on an additional vesicle sample of a narrower density interval (1.17–1.18 g ml-1). Separation by SDS-PAGE resolved a series of 32 bands that were isolated and subjected to nanoflow LC FT-ICR MS/MS analysis. From this sample, 126 different H. pylori proteins were identified. Among these, 93% were identical to the previous analysis and 7% were specific for MS II.

Mechanisms of Infection pathogenesis:

H. pylori is able to colonize and persist in a unique biological niche within the gastric lumen. The putative pathogenic determinants of H. pylori can be divided into two major groups’ virulence factors, which contribute to the pathogenetic effects of the bacterium, and maintenance factors, which allow the bacterium to colonize and remain within the host. H. pylori has three major pathogenic effects, gastric inflammation, disruption of the gastric mucosal barrier, and alteration of gastric physiology.

Induction of gastric inflammation:

(i) Interleukin-8: Interleukin-8 (IL-8) is a small peptide (chemokine) secreted by a variety of cell types, which serves as a potent inflammatory mediator recruiting and activating neutrophils. Several studies have
demonstrated that *H. pylori* strains are capable of inducing IL-8 secretion from gastric carcinoma cells in vitro. Wild-type strains that were VacA1 CagA1 produced significantly more IL-8 than did wild-type VacA2 CagA2 strains [38, 50, 78].

(ii) **Neutrophil adherence:** The 150-kDa proteins which increases the expression of neutrophil CD11b/CD18 and increases neutrophil adherence to endothelial cells have been characterized. The protein designated HP-NAP, is a polymer of 10 identical subunits. The gene (*napA*) shows homology to the gene encoding the bacterio ferritin family of proteins [114].

(iii) **Platelet-activating factor:** Platelet-activating factor (PAF) is a phospholipid mediator which is recognized as a potent ulcerogenic agent. Lyso-PAF is produced by gastric mucosal cells under basal conditions and in response to gastrin in healthy persons [298]. PAF stimulates gastric acid secretion via specific parietal cell receptors *H. pylori* can metabolize the non ulcerogenic precursor lyso-PAF into PAF. Thus, through synthesis of PAF, *H. pylori* may induce mucosal injury directly or indirectly via increased acid secretion [81].

(iv) **Lipopolysaccharide:** *H. pylori* LPS disrupts the gastric mucus coat by interfering with the interaction between mucus and its mucosal receptor; the anti-ulcer agent ebrotidine counteracts this effect [279]. However, the outstanding feature of the *H. pylori* LPS is its low proinflammatory activity [71]. This phenomenon is mediated by its unique lipid structure and affects binding to CD-14 [54].

(v) **Urease:** *H. pylori* urease is a potent stimulus of mononuclear phagocyte activation and inflammatory cytokine production [146]. In vitro, urease activity also is toxic to human gastric epithelial cells [80]. Thus, urease appears to function as both a colonization [maintenance] factor and a virulence factor. There is evidence that urease is associated with the outer membrane of *H. pylori*.

**Disruption of the gastric mucosal barrier:**

*H. pylori* can inhibit the secretory response of mucus cells in vitro, indicating a potential deleterious effect on this primary defense mechanism of the gastric mucosa [69].

(i) **Phospholipase:** *H. pylori* disrupts the protective phospholipid-rich layer at the apical membrane of mucus cells [61]. Furthermore, changes can be induced in phospholipid layers in vitro by phospholipases A2 and C expressed by *H. pylori* [75]; their effects can be inhibited by bismuth salts [75].

(ii) **Mucinase:** *H. pylori* possesses a gene that is almost identical to a mucinase gene of *Vibrio cholera* [79]. Such mucinase activity, if expressed in vivo, would probably contribute to disruption of the gastric mucosal barrier.

(iii) **Vacuolating cytotoxin:** Almost one-half of *H. pylori* strains produce a vacuolating cytotoxin in vitro [55]. This cytotoxin induces acidic vacuoles in the cytoplasm of eukaryotic cells; these vacuoles accumulate neutral lipids.
(iv) Reactive oxygen species: *H. pylori* induces the synthesis of reactive oxygen species (ROS) in gastric mucosa in vivo. There is a positive association between the amount of ROS present, the infective load of *H. pylori*, and the extent of gastric mucosal injury [39]. Smoking, drugs, and alcohol consumption had no independent effect upon ROS production in vivo [40]. There is no evidence for ROS participation in gastric mucosal injury in cases not related to *H. pylori* infection [40]. *H. pylori* stimulates mucosal ROS production both in vitro [29] and in vivo; this phenomenon appears to be of pathogenic significance [39].

(v) Inducible nitric oxide synthase: High-output nitric oxide production by inducible nitric oxide synthase (iNOS) is associated with immune activation and tissue injury. *H. pylori* induces iNOS in macrophages in vitro [335]. Eradication of *H. pylori* reduces iNOS in human gastric epithelial cells, suggesting that *H. pylori* also, induces the activity of this enzyme in vivo [58].

(vi) Apoptosis: *H. pylori* appears to induce programmed cell death (apoptosis) of gastric epithelial cells [58] and to stimulate oxidative DNA damage in infected human gastric mucosa [21]. In addition, *H. pylori* appears to inhibit gastric epithelial cell migration and proliferation [77]. Thus, *H. pylori* infection can induce gastric mucosal injury both directly and indirectly.

Altered gastric homeostasis:

*H. pylori* infection reversibly induces expression of the acid-stimulating peptide gastrin and suppresses expression of the acid-inhibitory hormone somatostatin [35]. These effects may not be due to *H. pylori* but may be related to the degree of gastric inflammation present. *H. pylori* increases the duodenal acid load under some circumstances but appears to decrease acid secretion under other conditions [85]. When acid secretion is suppressed with omeprazole, the relative susceptibility of *H. pylori* in the antrum and corpus differs, yielding the appearance of migration of bacteria from the antrum to the corpus [56].

(i) Motility: Motility is an essential colonization factor based on infection inability of the aflagellate, nonmotile variants of *H. pylori* [45]. Normally, *H. pylori* possesses two to six polar, sheathed flagella, whose filaments consist of two types, encoded by the flaAand flaB genes [53]. These genes have been cloned; the use of induced mutations has shown that both are essential for full motility [53] and for colonization [46]. The gene (flbA) that codes for the *H. pylori* homolog of a family of conserved proteins (LcrDInvA-FlbF family), which is involved in the regulation of motility, has been identified and cloned [83]. Mutants with mutations in the flbA gene are nonmotile and fail to express either flagellin or the hook protein [83].

(ii) Urease: All *H. pylori* isolates, as well as each of the gastric *Helicobacter* species, produce large quantities of the enzyme urease. There is evidence that urease is associated with the outer membrane of *H. pylori*. It is generally assumed that urease activity is required for production of a neutral microenvironment for the organism within the gastric lumen. [42].
(iii) Catalase and superoxide dismutase: The genes encoding the superoxide dismutase and catalase enzymes of *H. pylori* demonstrate significant homology to those of intracellular pathogenic microorganisms [82], suggesting a role in resistance to killing by polymorphonuclear leukocytes. Of interest, a significant fraction of catalase [76] and superoxide dismutase [82] is associated with the surface of viable *H. pylori*; whether surface association of these enzymes is essential for protection against oxygen-dependent killing of *H. pylori* by neutrophils is not known.

(iv) Heat shock protein homologs: The sequence of the gene encoding the HspB protein of *H. pylori* is highly conserved compared with those of heat shock proteins of other bacteria and humans [57]. This conservation suggests that the sequence cannot be modified without affecting the function of the protein. Based on structural similarity, HspB may function as a molecular chaperone for urease [44]. The gene (hspB) is part of a bicistronic operon (hspa-hspb), which has been cloned and sequenced. The hspa gene, which is located upstream of hspb, codes for the *H. pylori* homolog of the GroES heat shock protein homolog. The *H. pylori* hspa gene is unique in that it contains a nickel-binding site at its C-terminus. Expression of the hspa and hspb heat shock proteins together with the *H. pylori* urease increases the activity of urease in functional complementation experiments. Thus, hspa may play a role in the integration of nickel into the functional urease molecule [84].

(v) P-type ATPase: ATPases are of particular interest in *H. pylori* research, because an ATPase is presumed to be the target of the bactericidal action of proton pump inhibitors, such as lansoprazole or omeprazole, on *H. pylori* [72]. AP-type ATPase has been cloned and sequenced [63]. Isogenic mutants lacking this gene were viable; inactivation of the ATPase gene had no effect on the MIC of omeprazole, suggesting that this ATPase is not the target of the bactericidal action of omeprazole [127].

(vi) Siderophores: Iron is an essential element for bacterial growth and metabolism. *H. pylori* does not produce siderophores, (iron scavenging proteins) under iron-limiting conditions. However, *H. pylori* could grow in the presence of human lactoferrin and has surface receptors for this iron-binding protein [51]. In contrast, the production of siderophores was detected but did not affect the binding of lactoferrin to *H. pylori* proteins [52]. Thus, it is not yet clear how *H. pylori* acquires iron for its growth.

**Diagnosis and treatment:**

**Diagnosis of infection:** A variety of tests are now available to diagnose *H. pylori* infection. Histological examination of gastric tissue, bacterial culture, rapid urease testing, use of DNA probes, and PCR analysis, when used to test gastric tissue, all require endoscopy; therefore they incur expense and a risk of complication due to the procedure. In contrast, breath tests, serology, gastric juice PCR, and urinary excretion of ammonia are non-invasive tests that do not require endoscopy. The choice of test used for diagnosis of *H. pylori* infection will depend, in most cases, on the clinical information sought and the local availability and cost of individual tests [25].

**Treatment:** Although *H. pylori* is sensitive to a wide range of antibiotics in vitro, they all fail as monotherapy in vivo. In infected patients, the most effective single drug is clarithromycin, which leads to an approximate eradication rate of 40% when given twice daily for 10 to 14 days [14]. The lack of efficacy of monotherapy is related to the niche of *H. pylori*, residing at lower pH in a viscous mucus layer. Dual therapies, combining twice-daily-dosed PPI with, in particular, amoxicillin, are still in use in some countries, but dual...
therapies have mostly been replaced by triple therapies. These combine two antibiotics with either a bismuth compound or a PPI. A further alternative is provided by quadruple therapies, which combine the bismuth compound and PPI with two antibiotics. The exact mode of action of bismuth compounds is unknown, but Helicobacter pylori is susceptible to these compounds both in vivo and in vitro [11]. Tetracycline, amoxicillin, imidazoles [predominantly metronidazole and tinidazole], and a few selected macrolides [in particular clarithromycin, sometimes azithromycin] are probably the drugs most widely used for Helicobacter pylori eradication therapy recently, the use of rifabutin [19] and furazolidone [7] has been promoted. However, as their effectiveness is limited and many patients do not tolerate furazolidone, the primary use of these two antibiotics is a second-line rescue therapy of patients harboring metronidazole-resistant isolates [18].

REFERENCES


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