REVIEW ARTICLE

Gastric Pathophysiological Ins and Outs of Helicobacter pylori: A review

Jibran Sualeh Muhammad,[1] Toshiro Sugiyama,[2] Syed Faisal Zaidi[3]

Abstract

Helicobacter pylori infection induces chronic gastritis, peptic ulcer disease, gastric cancer and a number of related extragastric morbidities. Hence, it is now recognised as a worldwide problem. Although clinical outcomes are dependent upon bacterial virulence factors, host genetic diversity and environment, but the major focus of this review is on recent findings relevant to bacterial factors and gastric pathophysiology of H. pylori infection.

This article presents a review of the published literature mainly from year 2000 to 2012. The topics of main concerns were bacterial virulent factors and the inflammatory response to H. pylori infection. The authors used MeSH terms "Helicobacter" with "pathophysiology," "pathogenesis," or "gastric inflammation" to search the PubMed database. All relevant studies identified were included and are described according to the aforementioned subheadings.

Keywords: Helicobacter pylori, Pathogenesis, Pathophysiology, Inflammation.

Introduction

The gram-negative bacterium Helicobacter pylorus is the most common cause of chronic gastritis. It is highly adapted to the gastric environment where it survives within or beneath the gastric mucous layer. The bacterium renders the underlying gastric mucosa more exposed to acid peptic damage by disrupting the mucous layer, liberating enzymes and toxins, and adhering to the gastric epithelium (Figure-1). Also, it has been strongly linked to peptic ulcer disease and gastric cancer.[1] Therefore the pathophysiology of H. pylori infection and its eventual clinical outcome should be viewed as a complex interaction between the host and the bacterium. This interaction is influenced by the environment and modulated by a number of largely as yet unidentified factors.[2]

[1-3]Department of Gastroenterology and Hematology, Graduate School of Medicine and Pharmaceutical Sciences, University of Toyama, 2630 Sugitani, Toyama 930-0194, Japan, [1,3]Department of Biological and Biomedical Sciences, Aga Khan University, Karachi, Pakistan.

Correspondence: Syed Faisal Zaidi. Email: sfaisalhz@gmail.com

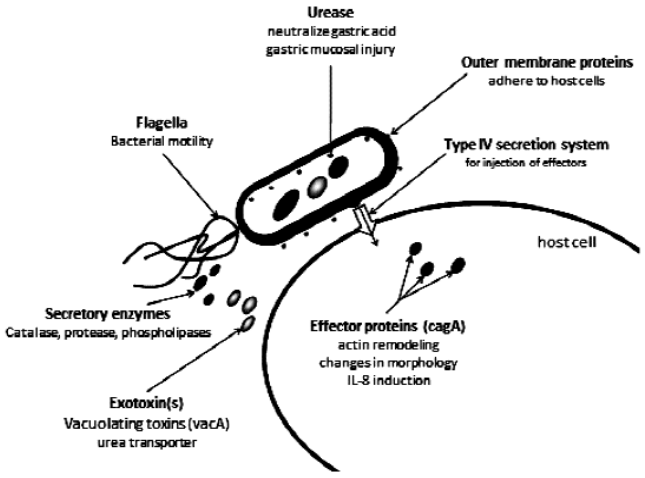

Figure-1: Bacterial factors responsible for virulence of Helicobacter pylori.

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Bacterial Factors

Tissue injury induced by H. pylori depends upon bacterial attachment and the subsequent release of enzymes and other microbial products that can cause cellular damage.

Bacterial attachment

H. pylori exclusively colonise gastric epithelium, suggesting the specific recognition of cell type by the bacterium. Numerous strategies to enable its survival and persistence within the gastric mucosa, including protein glycosylation, have been revealed recently. The presence of tight adherence of H. pylori to the gastric epithelial cell surface through formation of membrane attachment pedestals requires bacterial adhesins to recognise and specifically bind to host receptors expressed on the cell surface.[3] The attachment process alters the epithelial cell (morphologically or functionally) or activate certain

bacterial functions making them more toxic. At the site of adherence bacterial membrane proteins, coded by genes contained in the Cytotoxin-associated gene (Cag) pathogenicity island (PAI), open channels in the epithelial cell membrane that enable a direct contact of bacterial factors with the cell cytoplasm.[4]

Bacterial attachment is partially mediated by a number of adhesins and outer membrane proteins.[5] Blood group antigen binding adhesion (BabA), the best characterised of the three adhesin proteins, mediates binding to carbohydrate moiety of the fucosylated Lewis b [Le (b)] blood group antigens on host cells. Outer inflammatory protien (OipA) may not only serve as an adhesin but also triggers release of pro-inflammatory cytokines such as IL-8.[6] Sialic acid-binding adhesion (SabA) mediates binding to glycoconjugates containing sialyl Lewis receptors. Replacement of non-sialylated Lewis antigens by sialylated Le (x) or Le (a) has been associated with H. pylori induced gastric inflammation and cancer.[7]

Thus, the role of Lewis antigen expression in bacterial attachment is unclear. Nevertheless, the homologous structures of H. pylori lipopolysaccharide and host Lewis antigen may lead to an autoimmune response with subsequent cell injury.[8] H. pylori can also bind to class II major histocompatibility complex (MHC) molecules on the surface of gastric epithelial cells and induce apoptosis. In fact, binding of the organism's urease to surface class II MHC is itself sufficient to induce apoptosis.[9]

Release of enzymes

H. pylori secrete several enzymes that can cause cellular damage by direct or indirect mechanisms. Urea, when hydrolysed by bacterial urease, can form compounds such as ammonium chloride and monochloramine that can directly damage epithelial cells. It also alters the viscosity of gastric mucosa. In addition, the urease enzyme itself is antigenic and indirectly produces injury by stimulating inflammatory cells.[9]

Bacterial phospholipases can alter the phospholipid content of the gastric mucosal barrier, changing its surface tension, hydrophobicity, and permeability. The conversion of lecithin to lysolecithin (a toxic compound) by phospholipase A2 can lead to cell injury, while lipolysis can disrupt the structure and integrity of gastric mucus.[10]

H. pylori produces more catalase enzyme than most other bacteria. This enzyme, an antioxidant, may protect the organism from toxic oxygen metabolites liberated by activated neutrophils and allow it to survive and proliferate in an inflamed and damaged gastric mucosa. Hence, bacterial proteolytic enzyme activity can further degrade the mucus layer. However, the importance of proteolysis remains controversial.[10]

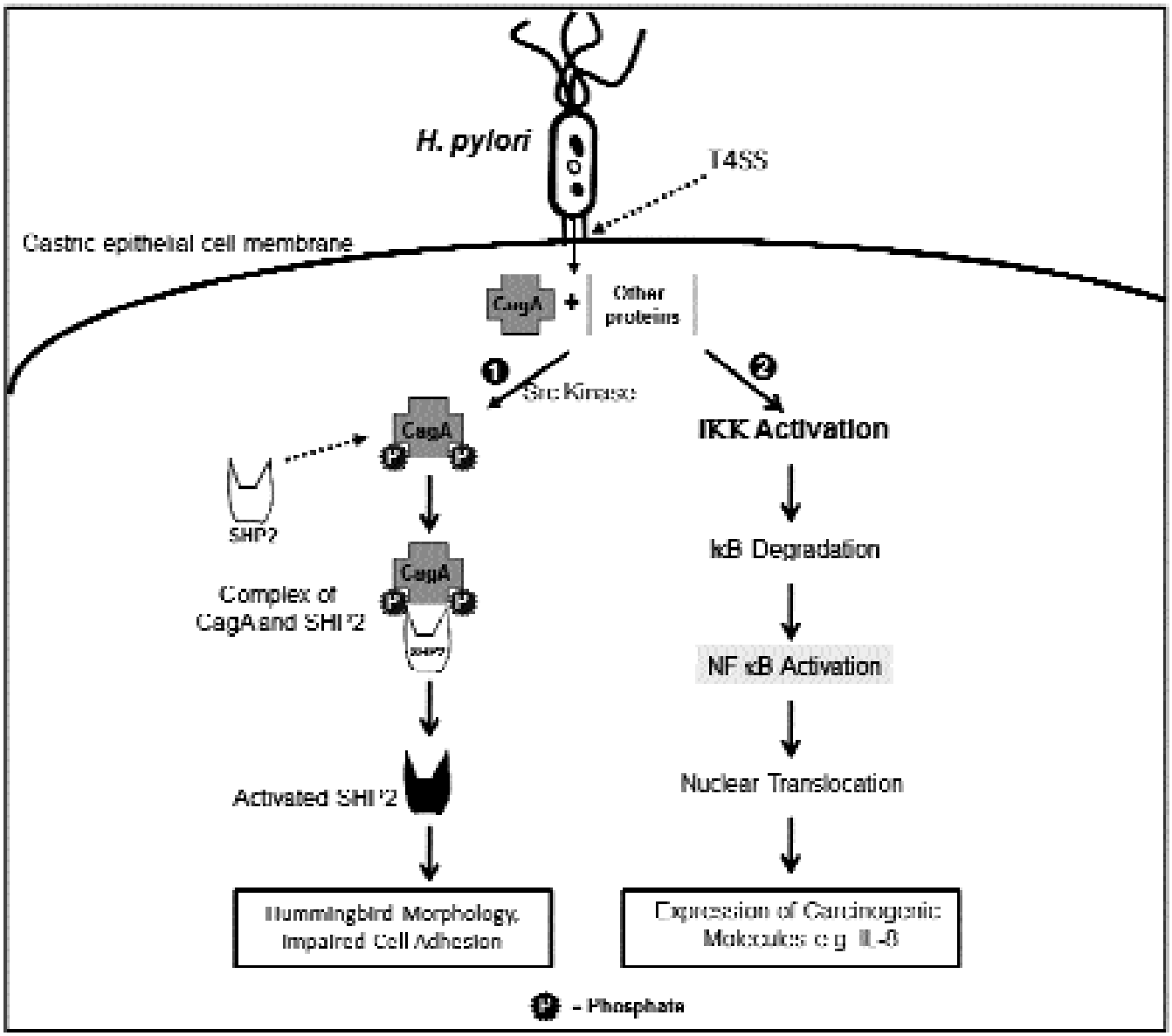
Bacterial strain differences

Functional differences exist between strains of H. pylori that may relate to virulence and tissue damage. One such difference is expression of an 87 kilodalton (kd) vacuolating cytotoxin (VacA) which causes cell injury in vitro and gastric tissue damage in vivo. All H. pylori contain the gene coding for VacA; however, only those strains that encode the Cag PAI, including CagA, coding for a 128 to 140 kd protein CagA, co-express VacA. VacA behaves as a passive urea transporter that is potentially capable of increasing the permeability of the gastric epithelium to urea, thereby creating a favorable environment for H. pylori infectivity.[11] Virulence of VacA appears to depend upon the function of a tyrosine phosphatase receptor in gastric epithelial cells.[12] H pylori strains with different VacA alleles have differing toxicity.[13]

CagA is not cytotoxic, but is antigenic and can be detected serologically. Its function is unknown, but, since it is necessary for VacA expression, it may play a role in transcription, excretion or function of the VacA cytotoxin. H. pylori can translocate its CagA protein into gastric epithelial cells via a type IV secretory apparatus. There it is tyrosine phosphorylated and possibly plays a role in host cell responses such as hummingbird morphology, actin re-modeling and impaired cell adhesion.[14-16]

Virulent strains of H. pylori encode Cag PAI, which expresses a type IV secretion system (T4SS). This T4SS forms a syringe-like pilus structure for the injection of virulence factors such as the CagA effector protein into host target cells. This is achieved by a number of T4SS proteins, including CagI, CagL, CagY and CagA, which by itself binds the host cell integrin member β(1) followed by delivery of CagA across the host cell membrane. A role of CagA interaction with phosphatidylserine has also been shown to be important for the injection process. After delivery, CagA becomes phosphorylated by oncogenic tyrosine kinases (e.g., Src Kinase) and mimics a host cell factor for the activation or inactivation of some specific intracellular signaling pathways i.e. protein tyrosine phosphatase pathway[14,15,17] (Figure-2).

Strains producing VacA and CagA cause more intense tissue inflammation and induce cytokine production.[18] Two other genes (PicA and CagE), which are co-transcribed and genetically linked to CagA, share a homology with genes coding for toxins in other known pathogenic bacteria. The gene product of CagE induces the release of epithelial cytokines, including IL-8.[19] This effect appears to be mediated by nuclear factor kappa B



(T4SS: type-4 secretion system, SHP2: Src Homology Phosphatases 2, IKK: I?B kinase complex, NF-?B: Nuclear factor kappa B, IL-8: Interleukin-8)

Figure-2: Important host cell intracellular pathways activated by Helicobacter pylori.

(NF- B), which activates transcription of IL-8 messenger ribonucleic acid (mRNA). In addition, CagA expressing bacteria are potent inducer of IL-8 promoter activity and secretion[20] (Figure-2).

The clinical significance of CagA positivity is demonstrated in two different disorders. Approximately 85-100% of patients with duodenal ulcers have CagA+ strains, compared to 30-60% of infected patients who do not develop ulcers. CagE positivity has also been associated with gastro-duodenal disease in adults and children.[21] CagA strains are also associated with a higher frequency of pre-cancerous lesions and gastric cancer. The risk of malignancy is thought to be related to a specific CagA motif and the intensity of CagA protein phosphorylation.[22] Yamada et al has demonstrated the presence of structural differences in CagA protein between the Western and Japanese bacterial strains, hence suggestting this difference as a possible linkage to the different disease outcome in Eastern patients.[23]

Other virulence factors

In addition to CagA, several other H. pylori virulence factors have been described.[24,25] The strength of these associations has not been well defined in large populations. Induced by contact with epithelium (IceA) has been associated with peptic ulcers.[24] Blood group antigen-binding adhesion (BabA2) has been associated with duodenal ulcers and gastric cancer. Outer inflammatory protein (OipA) has been associated with duodenal ulcers.[25]

Many of the virulence factors described above can coexist in the same H. pylori strain, making it unclear as to which factors might be most important. In addition, the expression of CagA is associated with both gastric cancer and duodenal ulcer, yet these two disorders rarely coexist. One study suggested that the OipA status may be a better predictor of H. pylori virulence than any of the other previously described virulence factors. The study included 247 patients infected with H. pylori (86 with gastritis, 86 with a duodenal ulcer, and 75 with gastric carcinoma), in whom H. pylori isolates were tested for other virulence factors discussed above. On multivariate analysis, only OipA status remained an independent predictor of H. pylori density, mucosal inflammation, and high mucosal IL-8 levels. However, the actual biological significance of these observations is unknown, and adaptation to gastric conditions is found to be mediated by phase variation of genes encoding for outer membrane proteins.[26]

Inflammatory Response

Although H. pylori is a non-invasive organism, it stimulates a robust inflammatory and immune response in the host cell. Various factors may contribute to these changes, which are described below. Bacterial colonisation, persistence and virulence, and resulting innate and adaptive host immune responses are all important in the pathogenesis of H. pylori related disease.[5,27,28]

The organism produces a number of antigenic substances, including heat shock protein, urease, and lipopolysaccharide, all of which can be taken up and processed by lamina propria macrophages and activate T-cells.[27] Cellular disruption, especially adjacent to epithelial tight junctions, undoubtedly enhances antigen presentation to the lamina propria and facilitates immune stimulation. The net result is increased production of inflammatory cytokines such as IL-1, IL-6, tumour necrosis factor alpha (TNF-), and most notably, IL-8.[28]

A B-cell response to H. pylori (with production of IgG and IgA antibodies) occurs locally in the gastroduodenal mucosa and systemically. The role of local antibodies in producing tissue injury or modulating inflammation in H. pylori infection remains controversial.[27] Prolonged stimulation of gastric B cells by activated T-cells can lead to mucosa-associated lymphoid tissue (MALT) lymphoma in rare cases.

T-cells are also activated during infection and their cytokines boost bacterial binding (by inducing class II MHC). While T-cells are recruited to the infected gastric mucosa, they appear to be hypo responsive. B7-H1 (programmed death-1 ligand 1); a member of B7 family of proteins associated with T-cell inhibition, appears to be involved in the suppression of T-cell proliferation and IL-2 synthesis during H. pylori infection, and thus may contribute to its chronicity.[29]

Different T helper cell subsets can be distinguished by their characteristic profiles of cytokine secretion. Th1 cells promote cell-mediated immune responses through elaboration of TNF-α and interferon (IFN-γ. T-helper (Th2) cells produce Il-4, IL-10 and TGF-b. It appears that during H. pylori infection the T-cell immunity is inappropriately skewed toward a Th1 response that promotes epithelial cell inflammatory cytokine production (IL-8 stimulated by IFN-γ and TNF-α) and directly impacts epithelial apoptosis.[30,31]

H. pylori infection induces a marked increase in the flux of leukocytes and in the appearance of platelet and leukocyte-platelet aggregates in gastric venules in a murine model. Circulating platelet aggregates and activated platelets were also detected in patients infected with H. pylori, suggesting that platelet activation and aggregation contribute to the associated micro-vascular dysfunction and inflammatory cell recruitment. Platelet aggregation mediated by an H. pylori interaction with von-Willebrand factor is speculated to contribute to infection related ulcer disease, but also possibly non-gastro-intestinal manifestations of infection such as cardiovascular disease and idiopathic thrombocytopenia.[32,33]

Not all H. pylori infected individuals develop clinical disease. Any number of host factors might contribute to the severity of inflammation by a wide variety of mechanisms.[34] Host genetics are important in determining the physiologic and clinical response to infection. Host IL-1 polymorphism determines the degree of inflammatory response to infection. Subsequently, it results in acid secretion alteration and risk for subsequent gastric cancer.[35] One series of meta-analyses investigated genes coding for the interleukin proteins (IL-1B, IL-1RN, IL-8, and IL-10) and for TNF-α. Gastric cancers were stratified

by histologic subtype and anatomic location, by H. pylori infection status, by geographic location (Asian or non-Asian study population), and by a quantitative index of study quality. Results consistently supported increased cancer risk for IL-1RN2 carriers; the increased risk was specific to non-Asian populations and was seen for intestinal and diffuse cancers, distal cancers, and, to a lesser extent, cardia region cancers. In Asian populations, reduced risk was observed in association with IL-1B-31C carrier status. These results indicate the importance of stratification by anatomic site, histologic type, H. pylori infection, and country of origin. Study quality considerations, both laboratory and epidemiologic, can also affect results and may explain, in part, the variability in results published to date.[36]

Interleukin-8

Research has centered on epithelial IL-8 production induced by different strains of H. pylori.[16] IL-8 is a potent chemotactic factor, activates neutrophils, and recruits acute inflammatory cells into the mucosa. H. pylori appear to activate transcription factor NF-κB via IκB kinase (IKK) pathway, which in turn increases IL-8 production.[20] NF-κB also regulates the expression of additional inflammatory response genes, and may play a role in the mucosal epithelial response to other bacterial infections in addition to H. pylori (Figure-2).

Bacteria that express CagA and VacA are more potent inducers of IL-8; however, the gene primarily responsible for IL-8 induction is CagE, which is located upstream of the CagA gene.[16] CagA/VacA-positive strains are also more often found in patients with clinical manifestations of H. pylori infection, indirectly suggesting that IL-8 may play an important pathophysiologic role in gastro-duodenal disease.

TNF-α can also augment IL-8 production by the inflamed mucosa. Following successful eradication of H. pylori, mucosal levels of mRNA for both TNF-α and IL-8 are reduced in parallel with the decline in local inflammation.[5]

Survival of H. pylori

H. pylori itself is in part able to survive this inflammatory onslaught by producing the enzyme, catalase. This enzyme neutralises the damaging reactive oxygen metabolites liberated by neutrophils.[9] With the passage of time, the host appears to down regulate the acute inflammatory response, making it easier for the organism to persist and proliferate.[7]

Antibody Response

Most infected individuals systemically produce specific antibodies to a variety of H. pylori antigens. The antibody response changes as infection progresses from an acute to a chronic stage. Detection of IgM antibodies is an insensitive indicator of acute infection and generally is clinically not useful, even in children. IgA and IgG antibodies are produced in response to infection, remain present as long as the infection is active, and quantitatively decrease after the infection is cured. Antibodies to CagA protein are detectable in gastric tissue and serum and permit the identification of infection with presumably more virulent organisms.[5]

The role of local antibodies in the immunopathogenesis of gastro-duodenal mucosal injury is unclear.[26] Virtually all infected persons have a specific gastric mucosal IgA and IgG response. IgA antibodies may modulate mucosal injury by inhibiting antigen uptake, disrupting bacterial adherence and motility, and neutralising various toxins. IgG presumably augments inflammatory injury by activating complement and facilitating neutrophil activation.

An antibody response may also be seen against autoantigens, including IL-8, antral epithelium and homologous host and bacterial epitopes (e.g., LewisX, lipopolysaccharide, and heat shock protein). The immunoglobulin specificity of MALT lymphoma may be for such autoantigens.[5,28]

Conclusion

The pathophysiology of H. pylori infection and its eventual clinical outcome should be viewed as a complex interaction between the host and the bacterium. This interaction is influenced by the environment and modulated by a number of largely as yet unidentified factors. Functional differences exist between strains of H. pylori that may relate to virulence and tissue damage. However, many of the virulence factors can coexist in the same H. pylori strains, making it unclear as to which factors might be most important. Although H. pylori is a non-invasive organism, but it stimulates a robust inflammatory and immune response. Bacterial colonisation, persistence and virulence, and resulting innate and adaptive host immune responses are all important in the pathogenesis of H. pylori related diseases. It is important to gain more insight into the pathogenesis of H. pylori-induced peptic ulcer disease and gastric cancer, not only to develop more effective treatments for these diseases, but also because it might serve as a paradigm for the role of chronic inflammation in the genesis of other clinical sequelae within and outside gastrointestinal tract.

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