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EDITORIAL

Roles of *Helicobacter pylori* BabA in gastroduodenal pathogenesis

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Abstract

Interactions between BabA and Lewis b (Le^b) related antigens are the best characterized adhesin-receptor interactions in *Helicobacter pylori* (*H pylori*). Several mechanisms for the regulation of BabA expression are predicted, including at both transcriptional and translational levels. The formation of chimeric proteins (*babA*/*B* or *babB*/*A* chimeras) seems to play an especially important role in translational regulation. Chimeric BabB/A protein had the potential to bind Le^b; however, protein production was subject to phase variation through slipped strand mispairing. The *babA* gene was cloned initially from strain CCUG17875, which contains a silent *babA1* gene and an expressed *babA2* gene. The sequence of these two genes differs only by the presence of a 10 bp deletion in the signal peptide sequence of *babA1* that eliminates its translational initiation codon. However, the *babA1* type deletion was found only in strain CCUG17875. A few studies evaluated BabA status by immunoblot and confirmed that BabA-positive status in Western strains was closely associated with severe clinical outcomes. BabA-positive status also was associated with the presence of other virulence factors (e.g. *cagA*-positive status and *vacA* s1 genotype). A small class of strains produced low levels of the BabA protein and lacked Le^b binding activity. These were more likely to be associated with increased mucosal inflammation and severe clinical outcomes than BabA-positive strains that exhibited Le^b binding activity. The underlying mechanism is unclear, and further studies will be necessary to investigate how the complex BabA-receptor network is functionally coordinated during the interaction of *H pylori* with the gastric mucosa.

Key words: *Helicobacter pylori*; BabA; Pathogenesis; Lewis antigens

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INTRODUCTION

The adherence of *Helicobacter pylori* (*H pylori*) to the gastric mucosa is widely assumed to play an important role in the initial colonization and long-term persistence in the human gastric mucosa. Analysis of the three completed *H pylori* genomes (strains 26695, J99, and HPAG1) has confirmed the presence of five major outer membrane protein (OMPs) families, which comprise approximately 4% of the *H pylori* genome. Among the families, members of the large Hop (*Helicobacter* outer membrane protein) family were the first characterized OMPs in *H pylori*. Several OMPs in the Hop family have been reported to act as adhesion molecules including the blood group antigen binding adhesin (BabA), sialic acid binding adhesin (SabA), adherence-associated lipoprotein (AlpA and AlpB), outer membrane inflammatory protein (OipA), and HopZ. Lewis b antigen (Le^b) and related fucosylated ABO blood group antigens are recognized by BabA[1], whereas sialyl-Lewis x and sialyl-Lewis a antigens (sLe^x and sLe^a) are recognized by SabA[2]. The corresponding receptors for AlpAB, OipA, and HopZ remain unknown. To date, BabA-Le^b is the best-characterized adhesin-receptor interaction in *H pylori*. In this review, I summarize recent data giving new insight into BabA and its role in pathogenesis.

IDENTIFICATION OF BABA

It is well known that Le^b is the dominant antigen in the

gastric mucosa of secretor-positive individuals[3], and the negative secretor status is associated with a Lewis a (Le^a)-dominant phenotype in the gastric mucosa (Figure 1). In 1993, two studies showed that *H pylori* can bind to fucosylated glycoconjugates containing Le^b structures on the surface of gastric epithelial cells within human biopsy specimens[4,5]. Studies using transgenic mice expressing the human Le^b epitope in gastric epithelial cells indicated that Le^b functions as a receptor for an *H pylori*-specific adhesin and mediates its attachment to the gastric pit and surface mucous cells[6]. Further studies using the same transgenic mice showed that *H pylori* was adherent to the surface of gastric epithelial cells, resulting in severe chronic gastric inflammation and atrophy; whereas *H pylori* was localized in the mucous layer in non-transgenic control mice[7].

In 1998, Ilver *et al* analyzed the blood group antigen-binding activity by measuring binding of *H pylori* to ^{125}I-labeled fucosylated blood group antigens[1]. Among 100 *H pylori* isolates examined, 66% bound the Le^b antigen; whereas 95% of the isolates did not bind the related Le^a, H-2, Le^x, or Le^Y antigens. The 78 K adhesin recognizing the Le^b antigen was detected on the bacterial cell outer membrane and was isolated by a combined ligand identification and purification technique and designated as blood group antigen-binding adhesin (BabA)[1]. Additional analyses revealed two sets of clones that encode two proteins with almost identical NH_2-terminal domains and completely identical COOH-terminal domains, but with divergent central domains. The corresponding genes were designated *babA* and *babB*; BabA but not BabB had Le^b antigen-binding activity. Therefore, the central domain in *babA* is believed to determine the specificity of receptor binding[1, 8-12]; however, the motifs of the *babA* gene that are involved in binding are still unknown.

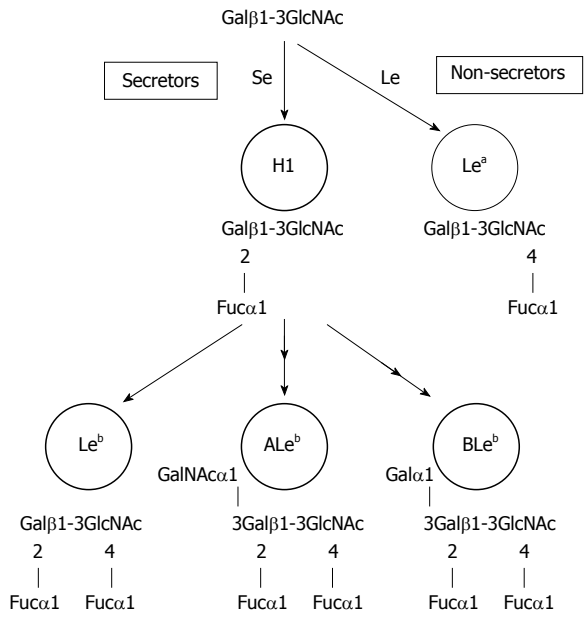

Figure 1 Biosynthetic pathways of Lewis antigens starting from type 1 lacto series core chains. Starting from type 1 core chains, an α1, 2-fucosyltransferase (Se) transfers fucose (Fuc) to the terminal galactose (Gal), resulting in the H-1 antigen (H1). H-1 antigen is a target for GalNAc- or Gal-transferases (in blood group A or B individuals) or remains unmodified (in blood group O individuals). These intermediates the are modified for the fucosylation step by an α1,3/4-fucosyltransferase (Le), resulting in the difucosylated histo-blood group antigens ALe^b, ALe^b and Le^b. Non-secretors are unable to produce an active Se product, and are only targets for the Le gene product Le^a.

FUNCTION OF BABA

BabA originally was defined as an adhesin binding to the Le^b antigen. The H-1 antigen is the carbohydrate structure that defines the blood group O phenotype in the ABO blood group system. Le^b, which is difucosylated, is formed by the addition of a branched fucose (Fuc) residue to H-1. The antigens that define blood group A and B phenotypes and corresponding antigens in the Lewis blood group system are formed by terminal N-acetylgalactosamine (GalNAc) or galactose (Gal) substitutions of H-1 and Le^b [A-1 and A-Lewis b (ALe^b), and B-1 and B-Lewis b (BLe^b) antigens, respectively; Figure 1].

Recently, Aspholm-Hurtig *et al* investigated the ability of BabA to bind Le^b, ALe^b and BLe^b[8]. Among 265 Le^b-binding *H pylori* strains from various geographic regions, more than 95% of *H pylori* strains are "generalists" (able to bind ALe^b and BLe^b in addition to Le^b); whereas a small subset of strains bind exclusively to ALe^b, and are called "specialist" strains. The authors proposed that the middle region of BabA was responsible for determining the different binding patterns; however, the specific motifs could not be identified[8]. Interestingly, "specialist" strains originated predominantly from South American individuals (where 60% of strains were classified as "specialist"), who are known to express almost entirely the blood group O phenotype. South American isolates in their study were from Peruvian and Venezuelan Amazon Amerindian populations and also from a Colombian mestizo (mixed Amerindian-European ancestry) population; probably most of these strains came mainly from European strains[13,14]. These data suggest that most specialist *babA* alleles may have arisen by mutation and/or recombination within the last 500 years. Therefore, the authors propose that such rapid evolution of BabA in response to host mucosal glycosylation patterns would enable the pathogen to adapt to their individual hosts while avoiding host immune responses, and contributes importantly to the extraordinary chronicity of human *H pylori* infection worldwide.

The mucins secreted by gastric mucous cells form a mucous gel layer covering the gastric mucosa. This gel layer is considered the first line of gastric mucosal defense against luminal noxious agents[15-17], and damage to the mucous gel is thought to precede gastric mucosal injury. The gastric surface mucous cells and gland mucous cells express the secretory mucins MUC5AC and MUC6/MUC5B, respectively[18,19]. The majority of *H pylori* reside in the gastric mucus overlying the epithelium. It is reported that *H pylori* could be co-localized with MUC5AC gastric mucin, but not with MUC6-producing cells in the glandular areas, suggesting

that adhesion is predominantly toward MUC5AC-specific ligands in gastric mucosa[20]. Subsequently, this binding phenotype could be correlated with the expression of an active BabA protein in *H pylori* and the presentation of the Le^b antigen in the gastric mucin MUC5AC[21,22]. However, since BabA-positive strains also attached to Le^b-negative MUC5AC of non-secretors, the involvement of additional epitopes and/or adhesins also must be involved[21]. In addition, binding of *H pylori* to MUC5B had been described[23] and a recent study confirmed that the binding was predominantly mediated by BabA and to a lesser degree by SabA adhesin[24].

LOCATION OF THE *BABA* GENE IN THE *H PYLORI* GENOME

H pylori 26695, J99 and HPAG1 each possess one *babA* allele (HP1243/JHP0833/HPAG1_0876) and one *babB* allele (HP0896/JHP1164/HPAG1_0320)[25-27]. Interestingly, the genomic locations of *babA* and *babB* genes are completely different among three strains (Figure 2). In strain J99, *babA* (JHP0833) is downstream of *hypD* (JHP0835) with a J99-specific gene (JHP0834) intervening, and *babB* (JHP1164) is downstream of *s18* (JHP1165). In strain 26 695, the locations of *babA* (HP1243) and *babB* (HP0317) are reversed. The chromosomal locations downstream of *hypD* and *s18* are referred to as locus A and locus B, respectively. In strain 26695, one gene encoding OMPs homologous to *babA* and *babB* (HP0317: denoted *babC*) with unknown function have been identified[27-29]. The location is referred to as locus C; interestingly, in strain HPAG1, the *babB* gene is located at locus C and *babC* gene at locus B. The *bab* genes initially were cloned from the strain CCUG17875, and this strain has two *babA* genes and one *babB* gene[1]. Gene inactivation experiments identified that only one gene (denoted *babA2*) had Le^b antigen-binding activity; whereas another gene (*babA1*) did not; *babA1* was located at locus B; however the locus of *babA2* was not determined[1].

The location of *babA* and *babB* in various clinical isolates of *H pylori* recently has been reported[29,30]. Hennig *et al*[30] analyzed a panel of 35 *H pylori* isolates and found that 24 (69%) contained *babA* sequences. In contrast, the *babB* gene was identified in 34 strains (97%). The *babA* gene was located at locus A for 19 strains (54%), at locus B for four strains (11%), and at locus C for three strains (9%). Four strains contain two copies of the *babA* gene, and the *babA* sequences found at two loci were identical in three strains and almost identical in one strain (i.e. three substitutions near the 5' ends of the genes in one strain), indicating that the multiple copies of *babA* presumably resulted from gene conversion (intragenomic nonreciprocal recombination) events. Importantly, two strains possessed the *babA* gene; however, the locus could not be identified, suggesting that there are additional unidentified chromosomal loci for *babA,* although *babA* may be found in one of three chromosomal loci in most cases.

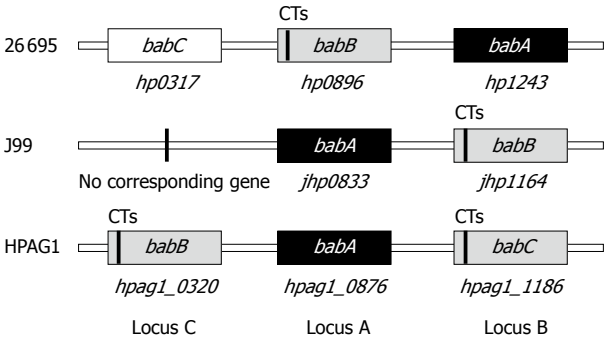

Figure 2 Genomic location of the *babA*, *babB*, and *babC* genes in strains J99, 26695, and HPAG1. CT: CT dinucleotide repeats.

Colbeck *et al*[29] analyzed a panel of 44 *H pylori* strains and found that 32 (73%) contained *babA* sequences. In contrast, the *babB* gene was identified in 41 (95%) isolates. The *babA* gene was located at locus A for 25 strains (57%) and at locus B for 18 strains (41%); locus C was not evaluated. Interestingly, although chromosomal DNA from low-passage-number, single-colony isolates was used, there was a mixed genotype in 30% (13/44) of the isolates, where the population of cells contained both *babA* and *babB* at the same locus. As a result, 11 strains were found to contain two copies of the *babA* gene including eight with mixed *babA* and *babB* at locus B.

Overall, from two detailed studies I conclude that the *babA* gene prefers to be located at locus A, some strains do not possess the *babA* gene, some strains possess multiple copies of the *babA* gene, and most strains possess the *babB* gene. The presence of *babB* might confer a stronger selective advantage than the presence of *babA*.

REGULATION OF BABA

Chimera formation between babA and babB

Several different mechanisms for regulation of BabA expression are predicted, including at the transcriptional and translational levels. As for translational regulation, the formation of chimeric proteins seems to play an important role. Chimera formation between *babA* and *babB* initially was reported by Pride and Blaser[11] who found that in two of 42 (5%) clinical isolates studied, the 5' regions of *babB* were replaced with the first 56 bp of *babA* (*babA/B* chimera). In addition, these authors showed that gene conversions frequently (10^{-3}) occur in *H pylori*, and the events are *recA*-dependent and DNase-resistant, indicating that they likely result from intragenomic recombination. *babA/B* chimeras also have been reported experimentally during *H pylori* infection in Rhesus monkeys[10].

In addition to *babA/B* chimeras, *babB/A* chimeras have been observed[9]. A *babA2* mutant from strain CCUG17875, defective in Le^b-binding, regained its activity by homologous recombination of a silent *babA1* gene into the *babB* locus, resulting in a chimeric *babB/A* gene. A silent wild-type *babA1* gene still was present.

The frequency of the *babA* mutant with Le^b-binding was approximately 10^{-5}. Detailed analyses of the chimeric *babB/babA* gene showed that the first 47 bp were *babB*-specific, the following 66 bp were shared between both *babA* and *babB*, and the remaining sequence was *babA*-specific. The second crossover event likely occurred within a region where the sequences of the *babA1* and the *babB* locus were identical. The chimeric BabB/A protein has the potential to bind Le^b; however, protein production was subject to phase variation through slipped-strand mispairing based on the number of Cysteine-Threonine (CT) dinucleotide repeats in the 5' region of the *babB* gene (switch "on" = functional and switch "off" = non-functional).

Initially, only five genes encoding the OMPs in *H pylori* (*oipA, sabA, sabB, babB* and *hopZ*) were reported to undergo phase variations in the 5' region such that not all strains produce functional proteins[25,27]. However, recent studies confirmed that phase variation is a method of regulating BabA production in some strains[10,29,30]. CT repeats were observed in 13 of 43 (30%) strains[29] and 4 of 22 (18%) strains[30]. Importantly, detailed analyses of the *babA* gene with CT repeats showed that the signal peptides are closely related to signal peptides of paralogous BabB proteins, whereas sequences further downstream were typical BabA sequences[30]. Taken together, these data suggest that the *babA* gene with the CT repeat might be the result of the translocation of *babA* into *babB* thereby generating a chimeric *babB/babA* gene. Interestingly, the *babC* gene in strain HPAG1 possessed CT repeats in the 5' coding region, whereas the *babC* gene in strain 26695 did not. These data suggest that *babB/C* chimera also might have occurred in some strains.

As described above, Colbeck *et al* found that there were cases with mixed *babA* and *babB* genes, especially at locus B[29]. The frequency of *babA* translocated at the *babB* locus was between 10^{-3} and 10^{-4}, which is in agreement with the frequency in strain CCUG17875[9]. Detailed investigation of 10 strains showed that the recombination event was identified from approximately 50 to 200 bp downstream of the ATG in five strains (all recombination occurred at locus B) and upstream of the ATG in the other five strains[29]. In the former case, the resulting gene forms the *babB/A* chimera, whereas complete recombination occurred in the latter case.

Overall, frequent translocation between *babA* and *babB* genes appears to be the main mechanism of regulating BabA expression. Therefore, *H pylori* uses both antigenic variation and phase variation to regulate *babA* expression.

Genomic mutations in the coding region of the babA gene

The *babA* gene initially was cloned from strain CCUG17875, which contains a silent *babA1* gene and an expressed *babA2* gene[1]. The sequence of these two genes differed only by the presence of a 10 bp deletion in the signal peptide sequence of *babA1* that eliminates its translational initiation codon. However, my group recently found that all 80 strains from a panel of Western and East Asian isolates contained an intact ATG start codon in the *babA* gene[31], and another group also reported the absence of the *babA1* type deletion[11,12,29,30,32]. Overall, the absence of a translation initiation codon, as described for *babA1* from CCUG17875, should be rare. Point mutations leading to stop codon, deletion and insertion in other parts of the *babA* gene also are not common; Hennig *et al* found one of 24 *babA*-positive strains (4%) contained a frameshift mutation that prevented expression of a full-length BabA protein (amino acid position at 55)[30].

Transcriptional regulation of BabA

Transcriptional regulation of BabA also has been reported. Backstrom *et al* found that only *babA2*, but not *babA1* was transcribed in strain CCUG17875[9]. Their analyses showed that *babA* transcription seemed to be regulated by the number of adenine [poly(A)] nucleotides within the -10 to -35 region of the *babA* promoter. The -10 and -35 region of the *babA2* sequences are highly homologous to the consensus for *E.coli* σ^{70} promoter sequences. This region was stable when the number of adenines was 10 (*babA2*) but would become non-functional when the number was 14 (*babA1*). The authors hypothesized that the poly(A) sequences between the -10 and the -35 sites could be prone to slippage mutations that allow changes in the level of transcription of downstream genes. However, other studies could not confirm that the -10 to -35 spacing played an important role in regulating *babA* expression[30,31]. Further studies will be necessary to fully interrogate the roles of transcriptional regulation of BabA.

Overall, there are several predicted mechanisms that may control BabA expression in some strains; however, there are many cases that remain unexplained. *H pylori* strains that do not produce BabA can be divided into five types, as shown in Table 1.

RELATIONSHIP BETWEEN BABA AND LE^B BINDING ACTIVITY

My group recently examined BabA protein and Le^b binding activity for 80 strains (40 from Japan and 40 from Colombia)[31]. BabA protein was measured by immunoblot analyses using anti-BabA antiserum (AK277), and Le^b binding activity was measured by binding of *H pylori* to ^{125}I-labeled fucosylated blood group antigens. *H pylori* strains were divided into two major groups: BabA-positive (76 strains) or BabA-negative (four strains). Semi-quantitative analyses of the BabA-positive strains allowed the BabA-positive strains to be classified into two distinct groups: those with high levels of BabA expression (68 strains) or those with low levels of BabA expression (eight strains). All of the 68 strains that exhibited Le^b binding activity produced high levels of BabA. The low and non-producer strains did not exhibit Le^b binding activity. Based on this finding, my group classified the strains into three distinct groups

Table 1 Five major types of *H pylori* strains that do not produce BabA

babA gene	Status
Negative	Include *babA/B* chimeras
Present	Regulated by slipped strand repairing and the status is "off" (probably equal to *babB/A* chimeras)
Present	Lack a translation initiation codon (single case of *babA1* in strain CCUG17875)
Present	Have a frameshift mutation(s) causing non-productive translation
Present	Without apparent mutations and without a hypothesis for the lack of expression

based on their expression levels of BabA: (1) BabA-high producers (BabA-H), which produce BabA protein at high enough levels to mediate Le^b binding, (2) BabA-low producers (BabA-L), which produce a small amount of BabA but not enough to mediate Le^b binding, and (3) BabA-negative strains, which do not produce any BabA protein.

BABA, LE^B BINDING ACTIVITY AND CLINICAL OUTCOMES

There currently are only a few studies that correlate the importance of BabA with clinical outcomes using immunoblot analyses[31,33,34]. My group recently performed large scale studies of 520 geographically diverse patients presenting with different clinical symptoms to evaluate BabA status by immunoblot analysis[31]. A total of 250 isolates from Western countries (150 strains from Colombia, 100 from the U.S.) and 270 isolates from East Asia (150 from Korea and 120 from Japan) were studied. All strains from East Asia expressed BabA protein. Twenty-four (9.8%) of Western strains were BabA-negative and were associated with milder gastric injury and lower *H pylori* density than BabA-positive status. BabA-negative status was inversely correlated with *cagA* status or *vacA* s1 genotype (i.e. only one (4.2%) and none (0%) of these BabA-negative strains were *cagA*- or *vacA* s1-positive, respectively). This is in agreement with previous studies that the *cagA* status was related to the presence of Le^b binding activity[1] and the presence of the *babA* gene[30].

Importantly, a small class of strains were BabA-positive but produced low levels of the BabA protein and lacked Le^b binding activity (BabA-L)[31]. Although these strains were functionally BabA-negative and were typically CagA-positive, they were more likely to be associated with duodenal ulcer, gastric cancer, and increased mucosal inflammation and atrophy than BabA-positive strains that exhibited *in vitro* Le^b binding activity (BabA-H strains) and BabA-negative strains. This finding suggests that either *in vitro* Le^b binding activity does not accurately reflect the severity of mucosal damage or that the clinical outcome or *in vitro* binding activity does not accurately reflect *in vivo* conditions. The underlying reason why strains with BabA-L status were more highly correlated with severe diseases than strains with BabA-H status is unknown, and it remains unclear whether expressing low levels of BabA have a direct role in the pathogenesis of gastroduodenal diseases. It is possible that BabA expression is influenced by the intragastric environment and that the phenotype of the BabA-L strains is an epiphenomenon rather than a cause of disease. It is possible that strong Le^b binding activity is associated with an inappropriate immune response resulting in severely inflamed mucosa. If so, the ability to change the BabA status from a high producer to low producer (i.e. Le^b binding to Le^b non-binding) would be advantageous for the organism, and a low producer might reflect an adaptation of *H pylori* that enhances survival in inflamed gastric mucosa. It also is possible that BabA expression down-regulates the proinflammatory effects of other putative virulence factors, such as the *cag* PAI and OipA.

DETECTION OF FUNCTIONAL *BABA* GENE

Most previous studies evaluating BabA (*babA*) status have used PCR techniques based on detection of the 10 bp deletion to distinguish between the *babA2* and *babA1* genes (Table 2)[35-53]. However, as described above, strains carrying the prototypical silent *babA1* gene are very rare, and in addition, the BabA protein levels often do not match the presence of the *babA* (*babA2*) gene[31]. Current terminology for *babA1* and *babA2* in the literature is confusing, and many researchers mistakenly understand that *H pylori* strains that do not produce BabA are either *babA* gene-negative or *babA1*-positive (= *babA* gene lacking a translation initiation codon). However, only one case with *babA1* has been reported, and BabA non-producing strains also usually possess non-functional silent *babA* gene sequences (i.e. 2, 4, and 5 in Table 2). Unfortunately, current PCR methods regard non-functional *babA* status as *babA2*-positive. In addition, a recent study confirmed that the PCR method used to detect *babA2* with only one primer pair previously designed yielded many false-negative results, probably due to sequence variation among strains[31].

Only a few studies have used a forward primer that is within the promoter region of the *babA* gene, a region that is identical to the sequence of *babA2* but different from that of *babA1* in strain CCUG17875[32,54,55]; however, recent analyses showed that the primers could also detect *babB* gene[31]. Overall, the information gained from currently used PCR-based methods must be interpreted with caution. In addition, I propose that researchers should not use current PCR-based methods in future studies.

Nonetheless, approximately half of the studies have suggested a correlation between *babA2*-positive *H pylori* in Western countries and increased risk of

Table 2 PCR-based genotyping for the *babA2* gene in *H pylori* positive cases *n* (%)

Study	Year	Population	Number studied	Prevalence of *babA2* gene						
				Total	Gastritis	PUD	Cancer	MALT	Duodenitis	Related to diseases
Western countries										
Gerhard *et al*	1999	Germany	114	82 (72)	18 (51)	23 (100)	21 (78)	20 (69)		Yes
Prinz *et al*[1,2]	2001	Germany	145	57 (39)	57 (39)					-
Rad *et al*[1]	2002	Germany	141	54 (38)	54 (38)					-
Zambon *et al*	2003	Italy	167	60 (36)	26 (28)	20 (49)			14 (42)	Yes
Oleastro *et al*	2003	Portugal	140	45 (32)	24 (23)	21 (58)				Yes
Podzorski *et al*	2003	USA	61	33 (36)	22 (36)					-
Oliveira *et al*	2003	Brazil	208	96 (46)	24 (32)	43 (54)	29 (56)			Yes
Rad *et al*[3]	2004	Germany	207	73 (35)	73 (35)					-
Lehours *et al*	2004	France	82	40 (49)	21 (54)			19 (44)		No
Gatti *et al*	2005	Brazil	89	42 (47)	37 (53)	3 (20)	1 (100)	1 (33)		No
Olfat *et al*	2005	Germany	92	41 (45)	19 (28)	22 (88)				Yes
		Sweden	74	33 (45)	21 (48)	12 (40)				No
		Portugal	91	31 (34)	12 (20)	19 (63)				Yes
		Finland	57	34 (60)	12 (46)	22 (71)				Borderline (P = 0.06)
Gatti *et al*	2006	Brazil	94	38 (40)	18 (41)	20 (40)				No
Asian countries										
Mizushima *et al*	2001	Japan	179	152 (85)	34 (81)	73 (85)	36 (90)	9 (82)		No
Yu *et al*	2002	China	104	83 (80)	83 (80)					-
Lai *et al*	2002	Taiwan	101	101 (100)	41 (100)	46 (100)	14 (100)			No
Han *et al*	2004	China	141	90 (64)	28 (65)	50 (65)	12 (57)			Yes (DU *vs* GU)
Zheng *et al*	2006	China	72	28 (39)	11 (39)	17 (40)				No
Lee *et al*	2006	Korea	135	83 (61)	64 (57)		19 (86)			Yes
Erzin *et al*	2006	Turkey	91	49 (54)	7 (23)	12 (43)	24 (73)			Yes

PUD: Peptic ulcer disease; MALT: Mucosal-associated lymphoid tissue; DU: Duodenal ulcer; GU: Gastric ulcer. [1]88% had German nationality and 12% were from other European countries; [2]Samples were examined from the antrum and the corpus, and the corpus data are presented (in the antrum, 55 were *babA2*-positive); [3]89% had German nationality and 11% were from other southern European countries.

developing significant clinical outcomes[38,44-46,52] and are in agreement with protein data as described above[31,33,34]. The prevalence of clinical isolates with a non-functional *babA2* gene without production of BabA protein may be low and negligible in some studies.

CONCLUSION

Several different mechanisms for regulation of BabA expression are predicted, including at both the transcriptional and translational levels. The formation of chimeric proteins seems to play an especially important role in translational regulation. The chimeric BabB/A protein has the potential to bind Le^b; however, the production was subject to phase variation through slipped-strand mispairing. Currently used PCR-based methods to evaluate BabA status do not take this mechanism of regulation into account, and information gained from currently used PCR-based methods must be interpreted with caution. I strongly recommend that researchers should not use PCR-based methods in their future studies. Recent studies evaluating BabA status by immunoblot confirmed that BabA-positive status in Western strains was closely associated with severe gastric injury, high *H pylori* density, and severe clinical outcomes. A small class of strains produced low levels of the BabA protein and lacked Le^b binding activity. Surprisingly, they were more likely to be associated with increased mucosal inflammation, atrophy, and severe clinical outcomes than BabA-positive strains that exhibit Le^b binding activity. The underlying reason is unclear, and further studies will be necessary to investigate how the complex BabA-receptor network is functionally coordinated during the interaction of *H pylori* with the gastric mucosa.

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