

## Prediction of Epitopes in the Proteome of *Helicobacter pylori*

Elkin Navarro-Quiroz<sup>1,2</sup>, Roberto Navarro-Quiroz<sup>3</sup>, Pierine España-Puccini<sup>2,4</sup>, José Luis Villarreal<sup>4,5</sup>, Anderson Díaz Perez<sup>1,9</sup>, Cecilia Fernandez Ponce<sup>6</sup>, Jorge Bilbao<sup>5</sup>, Lucy Vasquez<sup>7</sup> & Dary Luz Mendoza<sup>8</sup>

<sup>1</sup> Universidad Simón Bolívar, Barranquilla, Colombia

<sup>2</sup> E Biotechnology, Barranquilla, Colombia

<sup>3</sup> Centro de Investigación en Salud para el Trópico, Universidad Cooperativa de Colombia, Santa Marta, Colombia

<sup>4</sup> Universidad del Norte, Barranquilla, Colombia

<sup>5</sup> Universidad Libre, sede Barranquilla, Colombia

<sup>6</sup> Universidad de Cádiz, Cádiz, España

<sup>7</sup> Clínica la misericordia, Barranquilla, Colombia

<sup>8</sup> Universidad del Atlántico, Barranquilla, Colombia

<sup>9</sup> Universidad Rafael Nuñez, Cartagena, Colombia

Correspondence: Elkin Navarro-Quiroz, Profesor asistente, Universidad Simón Bolívar, Carrera 59 No. 59-65, Colombia. Tel: 300-532-4251. E-mail: enavarro26@unisimonbolivar.edu.co

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### Abstract

*Helicobacter pylori* (*H. pylori*) is classified by the World Health Organization (WHO) as a group I carcinogen and is one of the most efficient human pathogens with over half of the world's population colonized by this gram-negative spiral bacterium. *H. pylori* can cause a chronic infection in the stomach during early childhood that persists throughout life due to diverse mechanisms of immune response evasion. *H. pylori* has several factors strongly associated with increased risk of disease such as toxins, adhesins, and chemoattractants, some of which are highly polymorphic, phase variable, and have different functions. Conventional treatments involve the use of antibiotics. However, treatment frequently fails due to the resistance *H. pylori* has progressively developed to antibiotics. This creates the need for different treatments made possible by identifying new therapeutic targets in the pathogen's genome.

The purpose of this study was an *in silico* prediction of T- and B- epitopes in *H. pylori* proteins. Twenty-two external membrane proteins from *H. pylori* Strain 26695 (accession number NC\_000915) were identified using the web tool Vaxign (<http://www.violinet.org/vaxign/>). A total of one-hundred epitopes (60 class I epitopes and 40 class II epitopes) that could be used to develop novel non-antibiotics drugs for an *H. pylori* infection were predicted.

**Keywords:** *Helicobacter pylori*, epitopes, chronic infection in the stomach

### 1. Introducción

*Helicobacter pylori* (*H. pylori*) was first isolated by Warren and Marshall in 1982 (Ford & Axon, 2010). It is a slow-growing, Gram-negative, flagellated spiral-shaped microaerophilic organism (Jemilohun & Otegbayo, 2016). It has two to six flagella that provide mobility to withstand the rhythmic contractions in the stomach so they are able to penetrate the gastric mucosa (Spohn & Scarlato, 2001). It measures around 2.4 - 4.0 µm long and 0.5-1.0 µm wide (Refaeli et al., 2018).

*H. pylori* is an important pathogen in Public Health worldwide as one of the main causes of gastric diseases (Hooi et al., 2017). Several studies have shown that *H. pylori* is present in about 50% of the world population (Pilotto & Franceschi, 2014). In developing countries, the prevalence of infection can exceed 90% in adulthood (Hooi et al., 2017).

The host immune response induced by *H. pylori* is characterized by neutrophil recruitment followed by macrophages, mast cells, eosinophils, and T and B lymphocytes (Ayraud, Janvier, & Fauchère, 2002). Additionally,

there is an increase of IL-1, IL-8, and IL-6, and a reduction in the production of IL-17 (Kabir, 2011).

The current treatment of an *H. pylori* infection uses a combination of at least three compounds which include a proton pump inhibitor and two antibiotics. This treatment's decline in efficiency has been documented mainly due to its increased resistance to antibiotics (Fallone et al., 2016; Mégraud, 2012). Therefore, it is imperative to develop new therapeutic alternatives.

The rational design of a vaccine based on *In silico* prediction epitopes is a promising way to approach this problem (Moss et al., 2011). It has been demonstrated that surface proteins of some microorganisms have immunogenic epitopes, which could be used to design and develop novel, safe and efficient vaccines against pathogens (Ni, Wang, Liu, & Lu, 2010). However, the greatest requirement for the candidate epitopes is their ability to bind to molecules of Major Histocompatibility Complex (MHC) class I and / or class II. These molecules cause the epitopes to come to the cell surface to be subsequently recognition by the lymphocytes (Liao et al., 2009).

Vaxign is an online design system that predicts target proteins for vaccine design based on genome sequences using reverse vaccinology strategy (He, Xiang, & Mobley, 2010). The preestablished characteristics in Vaxign include the subcellular localization of the white protein and identifying epitopes that bind to MHC class I and class II (He et al., 2010; Xiang & He, 2009).

This study aimed to predict T- and B- epitopes from outer membrane proteins of *Helicobacter pylori* that can be used in later work to develop a vaccine against this microorganism or for alternative therapies such as passive immunization.

## 2. Methodology

### 2.1 Obtaining Genomic Sequences of *H. pylori*

The complete genome sequence of *H. pylori* was researched in RefSeq (Pruitt, Tatusova, & Maglott, 2005) and GenBank (Benson, Karsch-Mizrachi, Lipman, Ostell, & Wheeler, 2006) databases..

### 2.2 Identification of Conserved Sequences

The OrthoMCL algorithm was used to find conserved proteins (Yu et al., 2010) and SPAAN software (Sachdeva, Kumar, Jain, & Ramachandran, 2005).The transmembrane helix topology analysis was carried out using optimized HMMTOP (Xiang & He, 2009) is freely available to non-commercial users at <http://www.enzim.hu/hmmtop>. Source code is also available upon request to academic users.

### 2.3 Identification of Conserved Sequences

The OrthoMCL algorithm was used to find conserved proteins (Chen, Mackey, Stoeckert, & Roos, 2006), OrthoMCL provides a scalable method for constructing orthologous groups across multiple eukaryotic taxa, using a Markov Cluster algorithm to group (putative) orthologs and paralogs. This method performs similarly to the INPARANOID algorithm when applied to two genomes, but can be extended to cluster orthologs from multiple species. A cut off of E-105 was used as the default value for all proteins that could cross-react with human but mouse and/or pig proteins were discarded.

### 2.4 Prediction of Epitopes

For *H. pylori* protein, the Vaxitop pipeline was used for the prediction of epitopes (<http://www.violinet.org/vaxign/vaxitop>), following the methodology described above by (Xiang & He, 2013). Calculations were done using a cut-off point of  $p \leq 0.05$ , which provides high sensibility, specificity and balance (He et al., 2010).

## 3. Results

### 3.1 *H. pylori* Genomes

A total of 683 *H. pylori* genomes were found, of which only 61 sequences corresponded to genomes strains of this microorganism, with an average of 1,571 and 1,454 protein genes (Table 1).

Table 1. Genomes of *H. pylori* strains

Strain	Assembly	Genes	Proteínas
<i>Helicobacter pylori</i> 26695	GCA_000008525.1	1555	1445
<i>Helicobacter pylori</i> J99	GCA_000008785.1	1607	1469
<i>Helicobacter pylori</i> Shi470	GCA_000020245.1	1584	1442
<i>Helicobacter pylori</i> B38	GCA_000091345.1	1545	1432
<i>Helicobacter pylori</i> 51	GCA_000011725.1	1545	1433
<i>Helicobacter pylori</i> 52	GCA_000023805.1	1522	1392
<i>Helicobacter pylori</i> 908	GCA_000148665.1	1501	1360
<i>Helicobacter pylori</i> SJM180	GCA_000148855.1	1579	1488
<i>Helicobacter pylori</i> Cuz20	GCA_000148895.1	1580	1487
<i>Helicobacter pylori</i> 35 <sup>a</sup>	GCA_000178935.2	1530	1414
<i>Helicobacter pylori</i> India7	GCA_000185185.1	1602	1487
<i>Helicobacter pylori</i> F16	GCA_000270005.1	1531	1421
<i>Helicobacter pylori</i> F57	GCA_000270065.1	1560	1456
<i>Helicobacter pylori</i> 2017	GCA_000192315.1	1506	1382
<i>Helicobacter pylori</i> 2018	GCA_000192335.1	1516	1393
<i>Helicobacter pylori</i> 83	GCA_000213135.1	1562	1441
<i>Helicobacter pylori</i> Puno135	GCA_000224555.1	1585	1493
<i>Helicobacter pylori</i> Shi417	GCA_000277365.1	1586	1490
<i>Helicobacter pylori</i> Shi169	GCA_000277385.1	1559	1460
<i>Helicobacter pylori</i> Shi112	GCA_000277405.1	1607	1499
<i>Helicobacter pylori</i> PeCan18	GCA_000277425.1	1581	1480
<i>Helicobacter pylori</i> 26695	GCA_000307795.1	1632	1538
<i>Helicobacter pylori</i> Rif1	GCA_000307815.1	1630	1532
<i>Helicobacter pylori</i> Rif2	GCA_000307835.1	1630	1534
<i>Helicobacter pylori</i> OK113	GCA_000348865.1	1553	1418
<i>Helicobacter pylori</i> UM032	GCA_000392455.3	1549	1438
<i>Helicobacter pylori</i> UM299	GCA_000392475.3	1550	1439
<i>Helicobacter pylori</i> UM037	GCA_000392515.3	1634	1509
<i>Helicobacter pylori</i> UM066	GCA_000392535.3	1587	1474
<i>Helicobacter pylori</i> UM298	GCA_000439295.2	1551	1440
<i>Helicobacter pylori</i> SouthAfrica20	GCA_000590775.1	1552	1361
<i>Helicobacter pylori</i> 26695-1	GCA_000829095.1	1629	1539
<i>Helicobacter pylori</i> 26695-1CL	GCA_000829115.1	1628	1538
<i>Helicobacter pylori</i> 26695-1CH	GCA_000829135.1	1628	1538
<i>Helicobacter pylori</i> BM012A	GCA_000498315.1	1632	1495
<i>Helicobacter pylori</i> BM012S	GCA_000498335.1	1632	1495
<i>Helicobacter pylori</i> oki102	GCA_000600045.1	1566	1464
<i>Helicobacter pylori</i> oki112	GCA_000600085.1	1569	1456
<i>Helicobacter pylori</i> oki128	GCA_000600125.1	1496	1364
<i>Helicobacter pylori</i> oki154	GCA_000600145.1	1543	1412

Strain	Assembly	Genes	Proteínas
<i>Helicobacter pylori</i> oki422	GCA_000600165.1	1570	1441
<i>Helicobacter pylori</i> oki673	GCA_000600185.1	1534	1400
<i>Helicobacter pylori</i> oki828	GCA_000600205.1	1536	1399
<i>Helicobacter pylori</i> oki898	GCA_000600225.1	1563	1463
<i>Helicobacter pylori</i> J166	GCA_000685625.1	1569	1466
<i>Helicobacter pylori</i>	GCA_000685665.1	1551	1453
<i>Helicobacter pylori</i>	GCA_000685705.1	1626	1490
<i>Helicobacter pylori</i>	GCA_000685745.1	1551	1452
<i>Helicobacter pylori</i> NY40	GCA_000828955.1	1643	1476
<i>Helicobacter pylori</i>	GCA_000817025.1	1541	1413
<i>Helicobacter pylori</i> 26695-1	GCA_000826985.1	1630	1542
<i>Helicobacter pylori</i>	GCA_000827025.1	1631	1542
<i>Helicobacter pylori</i> J99	GCA_000982695.1	1594	1491
<i>Helicobacter pylori</i>	GCA_001433495.1	1617	1451
<i>Helicobacter pylori</i>	GCA_001653375.1	1584	1456
<i>Helicobacter pylori</i>	GCA_001653395.1	1573	1441
<i>Helicobacter pylori</i>	GCA_001653415.1	1590	1471
<i>Helicobacter pylori</i>	GCA_001653435.1	1462	1353
<i>Helicobacter pylori</i>	GCA_001653455.1	1498	1392
<i>Helicobacter pylori</i>	GCA_001653475.1	1491	1354

### 3.2 *H. pylori* Vaccine Candidate Protein Prediction

*H. pylori* strain 26695 was arbitrarily selected for the prediction of protein candidates. For this strain, a total of 1,445 proteins have been described. In this study, 42 outer membrane proteins were identified, with a sticky probability  $\geq 0.51$ . These proteins had no significant homology with pig, mouse or human proteins (Table 2). The outer membrane proteins with a greater likelihood adhesin (0.737) were: NP\_207714.1 (a protein with fusion functions) and NP\_207405.1 (a toxin-like protein).

Table 2. Selection of conserved outer membrane proteins of *Helicobacter pylori*, with probability of adherence  $\geq 0.51$

Protein Accession	Localization	Adhesin Probability
NP_206827.1	Outer Membrane	0.685
NP_207052.1	Outer Membrane	0.656
NP_207115.1	Outer Membrane	0.705
NP_207284.1	Outer Membrane	0.681
NP_207405.1	Outer Membrane	0.737
NP_207432.1	Outer Membrane	0.544
NP_207480.1	Outer Membrane	0.620
NP_207504.1	Outer Membrane	0.610
NP_207516.1	Outer Membrane	0.653
NP_207519.1	Outer Membrane	0.640
NP_207575.1	Outer Membrane	0.517

NP_207581.1	Outer Membrane	0.578
NP_207600.1	Outer Membrane	0.662
NP_207670.1	Outer Membrane	0.555
NP_207689.1	Outer Membrane	0.662
NP_207704.1	Outer Membrane	0.669
NP_207705.1	Outer Membrane	0.684
NP_207706.1	Outer Membrane	0.629
NP_207714.1	Outer Membrane	0.737
NP_208191.1	Outer Membrane	0.687
NP_208244.1	Outer Membrane	0.636
NP_208303.1	Outer Membrane	0.628

The outer membrane proteins with a greater likelihood adhesin (0.737) were: NP\_207714.1 (a protein with fusion functions) and NP\_207405.1 (a toxin-like protein).

#### Epitope Prediction of MHC Class I and II

Vaxitop (Xiang & He, 2013) was used to predict MHC class I and II binding epitopes. Likewise, other programs used to identify epitopes are listed in supplementary Table 1.

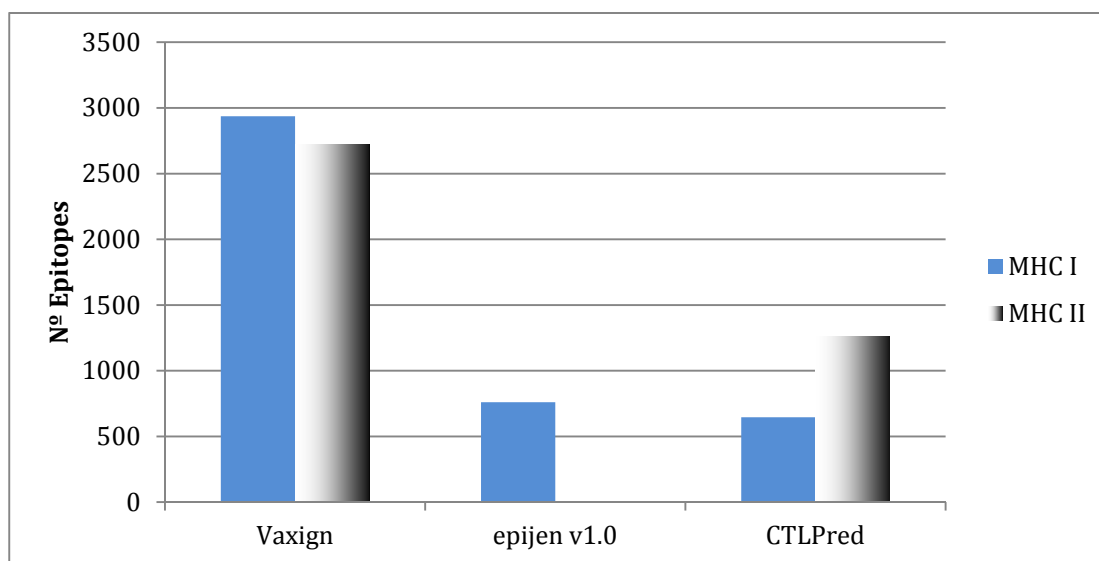


Figure 1. Total number of epitopes MHC I and MHC II predicted with different software

The greatest number of epitopes was obtained with Vaxign software (PSSM) with 2881 class I epitopes and 2634 class II epitopes. (Figure 1) The VacA protein (WP\_000874574.1), an *H. pylori* outer membrane protein toxin, presented the highest number of epitopes, as follows: 60 epitopes for MHC class I (Table 3), and 40 epitopes is for MHC class II (Table 4).

Table 3. Predicted epitopes for MHC I in proteins of *Helicobacter pylori*.

N° de acceso proteina	Epítotope	alelo MHC	aminoacido Inicial	aminoacido final	P-valor
WP_000726314.1	TTLNEACPWL	HLA-A*01:01	264	273	0.0159
	GTSWLNSQYV	HLA-A*02:01	619	628	0.0244
	LYSVYLNLYVF	HLA-A*24:02	700	709	6.50E-05
WP_000713712.1	LYSVYLNLYVF	HLA-A*24:02	734	743	6.50E-05
	SWLNSEYVNL	HLA-A*24:02	655	664	0.00732
	DTLVNFKSRY	HLA-A*01:01	479	488	0.00163
WP_001115603	QYRGFSWKIL	HLA-A*24:02	384	393	0.00022
	KALMVADLKY	HLA-A*01:01	233	242	0.0173
	ITYDTNPENF	HLA-A*01:01	267	276	0.0777
WP_000709741.1	RVKGLSIFYK	HLA-A*03:01	168	177	0.00044
	DYKRVS SVYL	HLA-A*24:02	286	295	0.0095
	LPYGFNTDLL	HLA-B*07:02	142	151	0.00792
WP_000822057.1	FIFDMMYTYK	HLA-A*03:01	645	654	0.00013
	PGLRYTFLNY	HLA-A*01:01	465	474	0.0247
	NVFGGVINVI	HLA-A*02:01	155	164	0.0184
WP_001228436.1	GYQNYFNDFI	HLA-A*24:02	508	517	0.0009
	QLTIENFLPY	HLA-A*01:01	95	104	0.0392
	FGDNLKTINL	HLA-A*01:01	333	342	0.0134
WP_001092132.1	KSDKAALGLY	HLA-A*01:01	79	88	0.00083
	YATGRFGNFY	HLA-A*01:01	332	341	0.018
	ATYRSNVANL	HLA-B*07:02	172	181	0.00582
WP_010875534.1	LYSVYLNLYVF	HLA-A*24:02	597	606	6.50E-05
	RLYSVYLNLYV	HLA-A*02:01	596	605	0.0002
	ITSTGPVTDY	HLA-A*01:01	203	212	0.00057
WP_000911476.1	FYSYGDKFHL	HLA-A*24:02	150	159	0.00018
	YVLYNSYLFY	HLA-A*01:01	142	151	0.00031
	VYRGFLWGIL	HLA-A*24:02	372	381	0.00063
WP_001108275.1	YMMDANAFTV	HLA-A*02:01	401	410	2.67E-05
	AYMQVDFTEL	HLA-A*24:02	67	76	0.00022
	FSADIKFEYY	HLA-A*01:01	452	461	0.00023
WP_010875548.1	FYFNYQRSYI	HLA-A*24:02	540	549	0.0002
	NTFKAYYQYY	HLA-A*01:01	275	284	0.00035
	QYNSYHPGTL	HLA-A*24:02	285	294	0.0016
WP_000479960.1	GLDYCGFDIY	HLA-A*01:01	677	686	0.00028
	DSLFEYGFNY	HLA-A*01:01	404	413	0.00613
	RPLRSNAIGL	HLA-B*07:02	257	266	0.0016
WP_000753173.1	FFDYNHAFIK	HLA-A*03:01	557	566	0.0174
	KQASIIITL	HLA-A*02:01	260	269	0.0244
	QELGRNPFRK	HLA-A*03:01	509	518	0.0715

WP_000592437.1	FYTKIGYKQF	HLA-A*24:02	350	359	0.00069
	LSTIGSQTNV	HLA-A*03:01	149	158	0.0477
	QTYSTQAIQY	HLA-A*03:01	206	215	0.0536
WP_000812546.1	VYLYMNSFL	HLA-A*24:02	197	206	0.00038
	NQLGNLIDLY	HLA-A*01:01	105	114	0.0159
	TYGVGTDVLY	HLA-A*01:01	410	419	0.0203
WP_001248496.1	MPRGNNTSYI	HLA-B*07:02	364	373	0.00061
	VTLAQVKVNL	HLA-A*01:01	62	71	0.0516
	MIMTTFPLYV	HLA-A*03:01	292	301	0.00064
WP_000874574.1	ALMSVSGQFV	HLA-A*02:01	1170	1179	0.00112
	GISGANGYEK	HLA-A*03:01	1214	1223	0.0193
	TTINLDDSVL	HLA-B*07:02	1097	1106	0.0241
WP_000902542.1	RYTYKDKFSF	HLA-A*24:02	605	614	0.00031
	AWCMTQHEGL	HLA-A*24:02	768	777	0.00732
	DHNVLTFFNY	HLA-A*01:01	564	573	0.00841
WP_000915379.1	YTNLSSQTNV	HLA-A*01:01	532	541	8.79E-05
	LPYNLNIEL	HLA-B*07:02	135	144	0.00483
	LPASLFNDPQ	HLA-B*07:02	170	179	0.0212
WP_000945747.1	YYNHQNIFYV	HLA-A*24:02	191	200	0.00057
	LSRFVTNMYV	HLA-A*01:01	773	782	0.00221
	TTKGERTFEV	HLA-A*01:01	32	41	0.00358

Table 4. Epitopes for MHC class II predicted by Vaxign for *Helicobacter pylori*

Nº de acceso	Epitopes	alelo MHC	aminoacido inicial	aminoacido final	Percentil (IEDB consensus)
WP_000726314.1	IQDNYIIDSNIHSQV	HLA-DRB1*04:01	496	510	0.01
	QDNYIIDSNIHSQVQ	HLA-DRB1*04:01	497	511	0.01
WP_000713712.1	ENIADTLVNFKSRYS	HLA-DRB1*13:01	475	489	0.02
	NIADTLVNFKSRVSE	HLA-DRB1*13:01	476	490	0.02
WP_001115603	YAIQRMVAPGINIT	HLA-DRB1*11:01	254	268	0.13
	FSNKYNIRMDLKLEY	HLA-DRB1*03:01	414	428	0.01
WP_000709741.1	LTPFNQVKSRTIFQL	HLA-DRB1*07:01	220	234	0.01
	NLTPFNQVKSRTIFQ	HLA-DRB1*07:01	219	233	0.01
WP_000822057.1	SFNTNYFVIFAKRYV	HLA-DRB1*13:01	556	570	0.01
	FNTNYFVIFAKRYVA	HLA-DRB1*13:01	557	571	0.01
WP_001228436.1	LINQNALPINYANLS	HLA-DRB1*13:01	437	451	1.52
	VATGLNYRYKHSKYS	HLA-DRB1*08:01	602	616	0.08
WP_001092132.1	QYYEYFNNLARMIA	HLA-DRB1*11:01	194	208	0.06
	QYYEYFNNLARMIAL	HLA-DRB1*11:01	195	209	0.06
WP_010875534.1	QLQYRRLYSVVLYNV	HLA-DRB1*01:01	591	605	0.09
	TQLQYRRLYSVVLYNV	HLA-DRB1*01:01	590	604	0.13

WP_000911476.1	GIKIHIDSNPKFRGL	HLA-DRB1*03:01	255	269	0.11
	EAPGIKIHIDSNPKF	HLA-DRB1*03:01	252	266	0.12
WP_001108275.1	VGYWGGLVGGQKPWAS	HLA-DRB1*13:01	124	138	0.11
	WGGLVGGQKPWASCGL	HLA-DRB1*13:01	127	141	0.11
WP_010875548.1	SVNASLQINNIFNMK	HLA-DRB1*13:01	749	763	0.07
	TSTDYFQIFNVMEGG	HLA-DRB1*13:01	562	576	0.45
WP_000479960.1	VFIKLDYTIPKTGI	HLA-DRB1*03:01	652	666	0.11
	IIKLDYTIPKTGINL	HLA-DRB1*03:01	654	668	0.28
WP_000753173.1	ANFQFLFNMGVVMNL	HLA-DRB1*11:01	641	655	0.61
	NFQFLFNMGVVMNLA	HLA-DRB1*15:01	642	656	0.88
WP_000592437.1	SQYLYSLLGAYPTKL	HLA-DRB1*01:01	82	96	0.05
	PTIYNTYYKSAGTTV	HLA-DRB1*07:01	485	499	0.2
WP_000812546.1	SQKVRFLAPLSLALS	HLA-DRB1*11:01	19	33	0.15
	QSQKVRFLAPLSLAL	HLA-DRB1*11:01	18	32	0.19
WP_001248496.1	EKYFLTSSLSLSLFL	HLA-DRB1*07:01	4	18	0.01
	FIDIVTLAQVKVNL	HLA-DRB1*11:01	58	72	0.34
WP_000874574.1	LLNFNGDRTLQNNAN	HLA-DRB1*03:01	999	1013	3.93
	QINTYTQMSRLAKL	HLA-DRB1*01:01	2200	2214	4.16
WP_000902542.1	HRVTGSLQINNIFNM	HLA-DRB1*13:01	798	812	0.07
	TGDPSFIKSLGNL	HLA-DRB1*07:01	217	231	0.27
WP_000915379.1	NLTNMLNMMAVFDS	HLA-DRB1*13:01	259	273	0.01
	VIYSYRVTNLYVNL	HLA-DRB1*07:01	342	356	0.24
WP_000945747.1	FEYNNKMYIDRKELQ	HLA-DRB1*03:01	39	53	0.1
	MYIDRKELQQRQSNQ	HLA-DRB1*03:01	45	59	0.24

## 5. Conclusions

In the present study, bioinformatic analysis was made to the complete genome of *H. pylori* (Strain 26695 with Gen Bank access number of NC\_000915). We identified 22 outer membrane proteins using the Vaxign web program. A total of 60 class I epitopes and 40 class II epitopes were predicted. These epitopes could be used in the improvement of active vaccines, for which we consider it important to combine diverse antigens of *H. pylori*, thus guiding the immune system to defend the host against this bacterium. Accordingly, this study was developed to predict antigenic determinants / epitopes of various outer membrane proteins of *H. pylori*. We propose probable epitopes of B and T cells that can trigger a desired immune response to these proteins. Additionally, they could be used in the design of therapeutic strategies without the use of antibiotics such as passive immunization, in which antibodies are administered to patients to help control infection by this microorganism, which we propose as the first line of action, before use of antibiotics.

## Competing Interests Statement

The authors declare that there are no competing or potential conflicts of interest.

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## Appendix

Table 1s. Comparison of four software for prediction of epitopes MHC I and MHC II classes of *Helicobacter pylori*

Accession #	software	prediccion petidos		metodo de predicion
		MHC I	MHC II	
WP_000726314.1	Vaxign	90	86	PSSM
	epijen v1.0	40		AGMP
	CTLPred	28		ANNs y SVMs
	imtech		3	MCTT y ANNs
WP_000713712.1	Vaxign	92	131	PSSM
	epijen v1.0	42		AGMP
	CTLPred	29		ANNs y SVMs
	imtech		3	MCTT y ANNs
WP_001115603	Vaxign	94	112	PSSM
	epijen v1.0	28		AGMP
	CTLPred	29		ANNs y SVMs
	imtech		51	MCTT y ANNs
WP_000709741.1	Vaxign	66	48	PSSM
	epijen v1.0	18		AGMP
	CTLPred	17		ANNs y SVMs
	imtech		34	MCTT y ANNs
WP_000822057.1	Vaxign	186	154	PSSM
	epijen v1.0	51		AGMP
	CTLPred	36		ANNs y SVMs
	imtech		4	MCTT y ANNs
WP_001228436.1	Vaxign	151	134	PSSM
	epijen v1.0	28		AGMP
	CTLPred	32		ANNs y SVMs
	imtech		68	MCTT y ANNs
WP_001092132	Vaxign	114	99	PSSM
	epijen v1.0	28		AGMP
	CTLPred	23		ANNs y SVMs
	imtech		51	MCTT y ANNs
WP_010875534.1	Vaxign	122	109	PSSM
	epijen v1.0	27		AGMP
	CTLPred	32		ANNs y SVMs
	imtech		61	MCTT y ANNs

WP_000911476.1	Vaxign	112	110	PSSM
	epijen v1.0	26		AGMP
	CTLPred	26		ANNs y SVMs
	imtech		48	MCTT y ANNs
WP_001108275.1	Vaxign	118	96	PSSM
	epijen v1.0	27		AGMP
	CTLPred	21		ANNs y SVMs
	imtech		52	MCTT y ANNs
WP_010875548.1	Vaxign	174	155	PSSM
	epijen v1.0	27		AGMP
	CTLPred	36		ANNs y SVMs
	imtech		81	MCTT y ANNs
WP_000479960.1	Vaxign	184	132	PSSM
	epijen v1.0	27		AGMP
	CTLPred	30		ANNs y SVMs
	imtech		82	MCTT y ANNs
WP_000753173.1	Vaxign	142	126	PSSM
	epijen v1.0	27		AGMP
	CTLPred	16		ANNs y SVMs
	imtech		73	MCTT y ANNs
WP_000592437.1	Vaxign	102	88	PSSM
	epijen v1.0	27		AGMP
	CTLPred	27		ANNs y SVMs
	imtech	55		MCTT y ANNs
WP_000812546.1	Vaxign	112	113	PSSM
	epijen v1.0	27		AGMP
	CTLPred	26		ANNs y SVMs
	imtech		57	MCTT y ANNs
WP_001248496.1	Vaxign	127	110	PSSM
	epijen v1.0	29		AGMP
	CTLPred	24		ANNs y SVMs
	imtech		55	MCTT y ANNs
WP_000874574.1	Vaxign	514	506	PSSM
	epijen v1.0	130		AGMP
	CTLPred	96		ANNs y SVMs
	imtech		249	MCTT y ANNs
WP_000902542.1	Vaxign	189	143	PSSM
	epijen v1.0	46		AGMP
	CTLPred	36		ANNs y SVMs
	imtech		89	MCTT y ANNs
WP_000915379.1	Vaxign	167	122	PSSM

	epijen v1.0	59		AGMP
	CTLPred	46		ANNs y SVMs
	imtech		112	MCTT y ANNs
	Vaxign	193	148	PSSM
WP_000945747.1	epijen v1.0	47		AGMP
	CTLPred	36		ANNs y SVMs
	imtech		89	MCTT y ANNs

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