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# IDENTIFICATION OF B CELLS EPITOPES FROM HSP60 GENES OF HELICOBACTER PYLORI FOR THE POTENTIAL DEVELOPMENT OF ENZYME IMMUNOASSAY

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**ABSTRACT:**
Helicobacter pylori (H. pylori), a microaerophilic, spiral-shaped, Gram-negative bacterium, a group I carcinogen since it infects the stomachs of over half of the world's population. Antigenic area or B-cell epitope refers to a portion or region of an antigen that is recognised by a particular antibody or B-cell. Aim of our study is to identify B cell epitomes from the HSP60 gene of Helicobacter pylori for the potential development of enzyme immunoassay. The amino acid sequence of the transmembrane protein of H.pylori was retrieved from the NCBI database and UniProt (ID FN598874). The sequence was utilized to identify potential immunogenicity B-cell epitopes using IEDB analysis resource online server program. The listed epitopes were tabulated and analyzed. Epitopes of length 15-25-mers were selected. Peptides "GYLSPYFVTNAEKMTAQ" and "VISEELGLSLENAEVEFLGK" are suitable for the potential development of B-cell epitope peptide vaccines and for the development indigenous antigen based enzyme immunoassay. Through this study we have identified peptide "GYLSPYFVTNAEKMTAQ"and "VISEELGLSLENAEVEFLGK" with the potential of peptide vaccine against H.pylori and for the development of specific enzyme linked assay.

**Keywords:** Helicobacter pylori, B cell epitope, enzyme linked immunoassay, hsp60, peptides, medicine, health, health system, health risk.

## 1. INTRODUCTION

The International Agency for Research on Cancer classified Helicobacter pylori (H. pylori), a microaerophilic, spiral-shaped, Gram-negative bacterium, a group I carcinogen since it infects the stomachs of over half of the world's population (IARC)(1). Chronic active gastritis, ulcers, lymphoma of the mucosa-associated lymphoid tissue (MALT), and stomach cancer are all strongly linked to H. pylori.(2) Additionally, if H. pylori infection is not treated, it usually develops in childhood and can last a lifetime (3). A triple or quadruple combination of antibiotics, bismuth drugs, and proton pump inhibitors forms the basis of the current therapy strategy (3,4). The downward trend of H. pylori eradication over the past ten years is due to an increase in clarithromycin resistance in many countries (5). The main causes of the failure of widespread control of H. pylori infection, in addition to the rising reports of antibiotic-resistant strains, are high treatment costs, subpar drug adherence, and a high rate of infection recurrence.(6) Research into immune defense mechanisms against H. pylori is therefore important for resolving issues with present treatment approaches and for improving the diagnosis, prevention, and treatment of H. pylori infection.

Antigenic area or B-cell epitope refers to a portion or region of an antigen that is recognised by a particular antibody or B-cell. There are two classes of these B-cell epitopes: continuous and discontinuous.(7) While a discontinuous/conformational epitope is a group of antigen residues that are far from one another in the main sequence but are brought to spatial proximity as a result of polypeptide folding,(8) a continuous/linear epitope is a stretch of consecutive residues in the primary sequence (9). Approximately 90% of B-cell epitopes are known to be conformational epitopes. (10)Clinical diagnosis assays utilizing synthetic peptides are thought to offer more benefits than those utilizing native or recombinant protein antigens. As a result, it's crucial to examine particular epitopes for the creation of epitope peptide-based diagnostic tools (11). B-cell epitopes are areas of the native antigen's surface that can be identified by specific antibodies or B-cell receptors.The labeled immunoassay known as the enzyme-linked immunosorbent assay, or ELISA, is the industry standard for immunoassays. This immunological test, which may identify and measure molecules like antibodies, antigens, proteins, glycoproteins, and hormones, is exceedingly sensitive. Antibodies and antigens are complexed in order to detect these products and produce a quantifiable result.

The immune system of a person creates a particular sort of protein called an antibody. This kind of protein has certain areas that bind to antigens. An antigen is a protein that can originate from a foreign substance and, when bound to an antibody, triggers an immune response in the body. With just a small amount of a test sample, this interaction can be used in ELISA testing to identify specific protein antibodies and antigens. Among other things, ELISA testing is used to detect HIV infection, conduct pregnancy tests, and type blood (12). Aim of the study is to identify the B cell epitopes from HSP60 genes of helicobacter pylori for the potential development of enzyme immunoassay.

## 2. MATERIALS AND METHODS:

The amino acid sequence of the transmembrane protein of H.pylori was retrieved from the NCBI database and UniProt (ID FN598874). The sequence was utilized to identify potential immunogenicity B-cell epitopes using IEDB analysis resource online server program. The listed epitopes were tabulated and analyzed. Epitopes of length 15-25-mers were selected

## 3. RESULTS:

In this study, a total of 20 peptides were identified of lengths varying from 1 to 35. Peptides lengths varying between 15 to 25 -mers were found to be effective. and more suitable for the potential development of B-cell epitope peptide vaccines and for the development indigenous antigen based enzyme immunoassay.

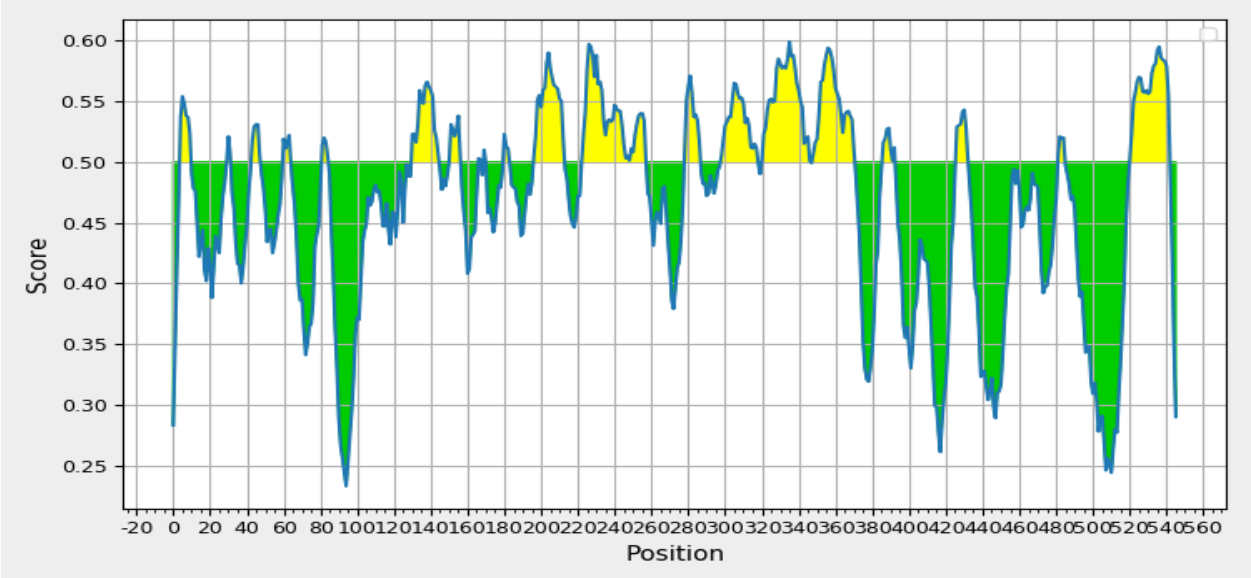


**Figure 1: B-cell Epitope Propensity Plot**

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| No | Start | End | Peptide | Length |
|---|---|---|---|---|
| 1 | 5 | 10 | IKFSDS | 6 |
| 2 | 31 | 32 | GP | 2 |
| 3 | 44 | 48 | GAPSI | 5 |
| 4 | 61 | 64 | LSCP | 4 |
| 5 | 82 | 85 | DAAG | 4 |
| 6 | 131 | 145 | KKASKKVGGKEEITQ | 15 |
| 7 | 152 | 157 | NSDHNI | 6 |
| 8 | 167 | 168 | KV | 2 |
| 9 | 170 | 170 | K | 1 |
| 10 | 181 | 183 | GIE | 3 |
| 11 | 197 | 213 | GYLSPYFVTNAEKMTAQ | 17 |
| 12 | 224 | 258 | KKISSMKDILPLLEKTMKEGKPLLIIAEDIEGEAL | 35 |
| 13 | 279 | 287 | GFGDRRKEM | 9 |
| 14 | 299 | 318 | VISEELGLSLENAEVEFLGK | 20 |
| 15 | 321 | 347 | RIVIDKDNTTIVDGKGHSHDVKDRVAQ | 27 |
| 16 | 349 | 371 | KTQIASTTSDYDKEKLQERLAKL | 23 |
| 17 | 387 | 393 | EMKEKKD | 7 |
| 18 | 427 | 433 | LNLHDDE | 7 |
| 19 | 482 | 485 | YVDM | 4 |
| 20 | 521 | 542 | IKEEKAAPAMPDMGGMGGMGGM | 22 |

**Figure 2: Tabulation of Predicted Peptides**

4. **DISCUSSION:** Hsps is one of the conservative proteins which are widely used proteins of the biosphere. For a long time, people have studied low Hsps as a molecular partner took part in physiological activities of cells. However, their results indicate that Hsps from microorganisms are the most important protective antigens when humans are infected with microorganisms (13). The protective immune response to 20 kinds of infectious diseases, such as tuberculosis and lepra was directly aimed at Hsps as far as reported. Especially in the model of mice infected with tubercle bacillus, specific anti-Hsp60 produced immunoreaction with Hsp60 of tubercle bacillus, not with mice Hsp60 themselves. This resolved the autoimmune problems because of high conservation. Furthermore, in mice and monkeys infected with plasmodium, immunoreactions did not depend on adjuvants when polypeptides were combined with Hsp60 (14). They showed that Hsp60 had similar functions of adjuvants. So under the uncertainty of *H pylori* urease preventing *H pylori* infection, *H pylori* Hsp60 as a *H pylori* vaccine component is not only used to combine urease to constitute multivalence vaccine, but also used as adjuvants to resolve the disadvantages of CT or LT.
   Previous studies shows that *H pylori* Hsp60 monoclonal antibodies significantly inhibits adhesion of *H pylori* to human gastric epithelial MKN45 cells and gastric cancer cells, also evaluated the protective effect of immunization with amino acids 189 to 203 (VEGMQFDRGYLSPYF) on *H pylori* Hsp60 molecules(15). The results suggested that the immune response to the epitope (VEGMQFDRGYLSPYF) could prevent *H pylori* infection in animal models. In this study, a specific antibody to Hsp60 was detected in sera from the mice immunized with purified recombinant protein Hsp60, but not in sera from the mice in the control group. These results suggest that Hsp60 of *H pylori* may be a good candidate for a vaccine and it can be used as an adjuvant and antigen.
5. **CONCLUSION:** The identification of the HspA protein fragments GYLSPYFVTNAEKMTAQ and VISEELGLSLENAEVEFLGKmarks a vital step toward a precision vaccine for *H. pylori*. As B-cell epitopes, these specific 17- to 20-mer sequences are the precise "keys" that the immune system's antibodies recognize to lock onto the bacteria. By focusing on these conserved segments of the Heat Shock Protein A, researchers can potentially train the body to neutralize the pathogen without the side effects often associated with whole-cell vaccines. While these peptides hold immense diagnostic and therapeutic promise, the next hurdle involves enhancing their stability and immune visibility through advanced delivery systems to ensure they provide lasting protection against gastric infection.

**FUTURE SCOPE:** With further investigations and knowledge on the B cell epitopes from hsp60 genes can be useful in formulating effective vaccines for Helicobacter pylori infections and diseases.

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**CONFLICTS OF INTEREST:**The authors declare that there are no conflicts of interest in the present study

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## 6. REFERENCES:

1. Arif M, Ahmad R, Sharaf M, Samreen, Muhammad J, Abdalla M, et al. Antibacterial and antibiofilm activity of mannose-modified chitosan/PMLA nanoparticles against multidrug-resistant Helicobacter pylori. Int J Biol Macromol. 2022 Nov 7;223(Pt A):418–32.
2. Website [Internet]. Available from: Mehta S, Rohinikumar S, Prabhu AR. Gene expression analysis of alkaline phosphatase in peri-implantitis tissue. 2023. https://pmc.ncbi.nlm.nih.gov/articles/PMC10563582/
3. Bai X, Zhu M, He Y, Wang T, Tian D, Shu J. The impacts of probiotics in eradication therapy of Helicobacter pylori. Arch Microbiol. 2022 Nov 7;204(12):692.
4. Kountouras J. Concentrating on the long topic of empirical or not conventional Helicobacter pylori eradication regimens. Clin Gastroenterol Hepatol [Internet]. 2022 Nov 4; Available from: http://dx.doi.org/10.1016/j.cgh.2022.10.027
5. Graham DY. Role of Helicobacter pylori in NSAID gastrophy: can H. pylori infection be beneficial? [Internet]. Helicobacter pylori. 2000. p. 453–9. Available from: http://dx.doi.org/10.1007/978-94-011-3927-4_48
6. Ann Preethy N, Somasundaram S. Safety and physiologic effects of intranasal midazolam and nitrous oxide inhalation based sedation in children visiting Saveetha Dental College and Hospitals, India. Bioinformation. 2022 Jan 31;18(1):26–35.
7. Sathiyamoorthy S, Gheena S, Jain RK. Prevalence of oral mucocele among outpatients at saveetha dental hospital, india. Bioinformation. 2020 Dec 31;16(12):1013–8.
8. Website [Internet]. Available from: Sureshbabu NM, Selvarasu K, Kumar JV. Concentrated Growth Factors as an Ingenious Biomaterial in Regeneration of Bony Defects after Periapical Surgery. 2019. https://pmc.ncbi.nlm.nih.gov/articles/PMC6362495/
9. Martineau P, Guillet JG, Leclerc C, Hofnung M. Expression of heterogonous peptides at two permissive sites of the MalE protein: antigen city and immunogenicity of foreign B-cell and T-cell epitomes [Internet]. Vol. 118, Gene. 1992. p. 151. Available from: http://dx.doi.org/10.1016/0378-1119(92)90266-r
10. Pradeep M, Balakrishnan N, Arvind TRP. Prevalence of usage of various removable appliances among undergraduate dental students in a private dental college. J Adv Pharm Technol Res. 2022 Dec;13(Suppl 2):S559–62.
11. Palatnik-de-Sousa CB, da Silva Soares I, Rosa DS. Epitope Discovery and Synthetic Vaccine Design. Frontiers Media SA; 2018. 284 p.
12. Hashim OH. The ELISA test [Internet]. Vol. 19, Biochemical Education. 1991. p. 38. Available from: http://dx.doi.org/10.1016/0307-4412(91)90146-y
13. Romano C. Commonly used antibiotics induce expression of Hsp 27 and Hsp 60 and protect human lymphocytes from apoptosis [Internet]. International Immunopharmacology. 2004. Available from: http://dx.doi.org/10.1016/s1567-5769(04)00143-2
14. Bai Y, Li LR, Wang JD, Chen Y, Jin JF, Zhang ZS, et al. Expression of Helicobacter pylori Hsp60 protein and its immunogenicity. World J Gastroenterol. 2003 Dec 12;9(12):2711.
15. Yamaguchi H, Osaki T, Kurihara N, Taguchi H, Kamiya S. Reactivity of monoclonal antibody to HSP60 homologue of Helicobacter pylori with human gastric epithelial cells and induction of IL-8 from these cells by purified H. pylori HSP60. J Gastroenterol. 1999;34 Suppl 11:1–5.