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Design of HIV-1 Peptide-Based Vaccine from Matrix Protein p17: Immunoinformatics Approach

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A R T I C L E I N F O

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A B S T R A K

HIV/AIDS infection is a spectrum of infectious diseases of the immune system. Rapid transmission causes difficulty in overcoming this disease. This studied aims to determine potential peptides as vaccine candidates, and to determine the physicochemical properties and homologous properties of vaccine candidates. This studied used a processing method with an immunoinformatics approached and used samples from the p17 matrix protein sequence with PDB code 2HMX. All stages were carried out used the appropriate web server and application. From sequence analysis, selected T-cell and B-cell epitopes was obtained. After designing the vaccine candidate from T-cell and B-cell epitopes, the final stepped was to perform 2D and 3D visualization of the vaccine candidate. The researched shows that the peptide were as follows, adjuvant-**EAAAK**-GSEELRSLY-**AAY**-NNSQVSQNY-**GPG PG**-TGSEELRSLYNTIAV-**KK**-QPSLQTGSEELRS -**KK**-DVKDTKEALDK. The physicochemical properties of the HIV vaccine candidate had a total of 116 amino acids, a molecular weight of 12871.66 g/mol, a theoretical pI of 9.58, a number of negative residues 13, a number of positively charged residues 24, the extinction coefficient was 7825 m^2/mol, the stability index was 59, 77, aliphatic index 64.74, average hydrophobicity -0.958. The results of the homologous analysis of the matrix protein p17 vaccine candidate stated that the amino acid residues that made up the peptide were not homologous or did not caused an autoimmune response when used as a vaccine candidate.

INTRODUCTION

The Human Immunodeficiency Virus (HIV) is a ribonucleic acid (RNA) virus that belongs to the retrovirus lentivirus genus. The virus enters the host's body and affects CD4 T lymphocyte cells in regulating and maintaining the immune system. This process causes continuous replication that eventually lysates lymphocyte cells. HIV remains a major challenge in the health world. Rapid transmission and development pose as one of the difficulties in dealing with the disease. Minimal knowledge amongst the community is one of the supporting factors for HIV to continue increasing every year.

Human immunodeficiency virus type 1 (HIV-1) is the leading cause of HIV disease, with the end stage being Acquired Immune Deficiency Syndrome (AIDS). AIDS is a major global public health problem. Antiretroviral therapy (ART) is one therapy that can inhibit HIV replication and improve patient prognosis, converting AIDS into a chronic viral infection. In most individuals, HIV-1 induces immune activity that involves not only the main infection targets (CD4 T lymphocytes and monocytes/macrophages) but also B lymphocytes, natural killer cells, and antigen presenting cells. The direct effect of HIV-1 gene products in triggering chronic immune stimulation is where virus proteins can create an active immunological environment.

Out of many HIV-1 proteins, matrix protein p17 is the target for antibody neutralizers against HIV-1. Matrix protein p17 serves several functions in the virus replication cycle. The function of matrix protein p17 is to recruit surface protein/transmembrane complex gp120/gp41 of the virus into the virus particle. Furthermore, matrix protein p17 functions as a protein that targets Pr55Gag protein to its assembly site on the infected plasma membrane.

This peptide-based vaccine design is a vaccine design composed of several amino acid residues, usually made up of 9-15 residues. Amino acids are the smallest part of a protein, where a protein is the main compound of peptide vaccine candidates. Using a peptide-based vaccine design can help monitor diseases and stimulate disease responses. The principle of this peptide-based vaccine design is based on the immune system's response to the antigen. The HIV vaccine is developed by substituting epitopes from the matrix protein p17. Epitope substitution is expected to provide an immune response to the HIV virus, thus reducing the mortality rate caused by HIV infection.

In this vaccine design research, an immunoinformatics approach is used to predict vaccine candidates from virus proteins. Considering there is still no treatment that can cure HIV/AIDS, as well as in terms of prevention, this research can simplify predicting vaccine candidates. This vaccine design using this method holds the potential to design vaccine candidates that can play an important role in disease diagnosis and management. The immunoinformatics method works by predicting peptide binding with MHC. The immunoinformatics method significantly reduces time, cost, and labor involved in experimental verification. Therefore, peptide-based vaccine design using computational methods is beneficial for vaccine development in Indonesia, especially for HIV.

This research designs peptides from the matrix protein p17 using appropriate web servers for testing. The characteristics of the produced epitopes are determined from antigenic, allergenic, toxic, and homologous properties. This research aims to find out the potential peptide sequence and position as a vaccine candidate, determine the physicochemical properties of the HIV vaccine candidate, and ascertain the homologous properties of the vaccine candidate. The predicted vaccine candidate results are expected to be useful for vaccine development in Indonesia.

LITERATURE REVIEW

Human Immunodeficiency Virus (HIV)

Human Immunodeficiency Virus (HIV) was a virus that caused Acquired Immunodeficiency Syndrome (AIDS). AIDS was a collection of symptoms resulting from the weakened immune system due to the entry of HIV virus into a person's body. HIV belonged to the retrovirus group, which was a type of virus that possessed an enzyme (protein) capable of converting its genetic material, RNA, into DNA. This group was called retroviruses because they reversed the normal sequence, transforming DNA into RNA. After infecting a host, HIV's RNA transformed into DNA through the action of the reverse transcriptase enzyme. The DNA was then inserted into human cells' DNA. This DNA could be used to create new viruses that infected new cells or remained dormant within long-lived cells or reservoirs, such as resting CD4 cells. HIV's ability to remain hidden was the reason why the virus persisted throughout a person's lifetime, even with effective treatment.

Structure of HIV

Currently, HIV was classified into two types: HIV type 1 and HIV type 2. Worldwide, the primary agent of AIDS was HIV-1, while HIV-2 was limited to certain regions of West and Central Africa. HIV was a genetically related member of the Lentivirus genus within the Retroviridae family. Lentivirus infections typically exhibited a chronic disease course with a long clinical period. HIV-1 and HIV-2 differed in their genomes, although their basic structures were similar.

Matrix Protein p17

Matrix protein p17 was a 132-amino acid polypeptide that formed the protective shell lining the inner side of the virus's plasma membrane. Matrix protein p17 played a key role in several steps during the virus replication, both in the early and late stages of the virus life cycle. The viral protein was continuously released into the extracellular space from HIV-1-infected cells and could be detected in the plasma of HIV-positive patients and in tissue specimens.

Vaccine

The term 'vaccination' was first used by Edward Jenner in 1796 to describe the injection of the smallpox vaccine. Vaccination involved the administration

of antigenic agents to stimulate an individual's immune system and develop adaptive immunity against a particular disease. Vaccines could improve or even prevent the effects of an infection. Vaccination was generally considered the most effective method for preventing infectious diseases, and its efficacy had been extensively studied and verified. Some vaccines were administered after a patient had been infected by a pathogen. Vaccination administered within the first three days after exposure to smallpox was reported to greatly weaken the disease, and vaccination given up to a week after exposure could provide protection from the disease or reduce its severity.

Immune System

The immune system was a collection of immune cells found in the blood, lymphatic fluid, tissues, and organs that worked together to protect the body against foreign substances such as microbes (bacteria, fungi, and parasites), viruses, cancer cells, and toxins. The immune system was highly complex, capable of recognizing and remembering millions of different foreign substances. Immunity or resistance to infections was acquired from the activities and functions of two closely related systems, namely the innate immune system and the adaptive immune system. The external defenses, such as the skin and mucous membranes, phagocytic leukocytes, and serum proteins, were included in the elements of the innate immune system, which worked non-specifically against foreign substances or cells without the need to recognize specific identities. The adaptive immune system, on the other hand, differed from the innate immune system and relied on specific recognition by lymphocytes of foreign substances or cells.

Antigen

There were two important characteristics of antigens: immunogenicity, the ability to elicit an immune response (either by stimulating the production of specific antibodies, T cell proliferation, or both), and reactivity, the ability of antigens to interact specifically with antibodies or cells. Antigens could encompass whole or parts of microbes, chemical components of bacterial structures, non-microbial chemical components (such as egg whites, pollen, tissues, or organ transplants). Certain small portions of large antigen molecules could trigger an immune response, referred to as epitopes or antigenic determinants.

Epitope was a structure or part that was immunogenic from a protein molecule or antigen. Epitopes could belong to foreign proteins or self-proteins, and they could be categorized as conformational or linear, depending on their structure and integration with paratopes. T cell epitopes were presented on the surface of antigen-presenting cells (APCs), where they bound to major histocompatibility complex (MHC) molecules to induce an immune response. MHC was located on the short arm of chromosome 6, making it the most complex genetic system in the human genome, and it included the HLA genes. Transmembrane HLA proteins encoded by classical HLA genes (A, B, C, DR, DQ, and DP) were primarily involved in antigen presentation on the cell surface, from small pathogen-derived peptide fragments to T cells, triggering

an immune response. Different HLA alleles presented different repertoires of peptide fragments from attacking pathogens, potentially influencing T cell immune responses.

Immunoinformatics

Immunoinformatics or computational immunology was a field that connected computer science and immunology, utilizing computational resources and methods to handle and understand immunological data. Informatics had been incorporated into many immunological topics, ranging from disease prevention and diagnosis to drug discovery. Current applications heavily relied on the interpretation of immunology laboratory reports. Results were obtained using computational methods, but many advancements in the field had already made it possible to be purely data-driven, with in silico discoveries being made using publicly available data.

METHODOLOGY

The tool used in this research was an Acer notebook with an Intel(R) Core(TM) i3-1115G4 CPU @ 3.00GHz (4 CPUs), ~3.0GHz, 4GB RAM, and Windows 10 operating system. This was a computational study that utilized data from an online database. The material used was a sample sequence of the matrix protein p17.

Study Design: Computational study

Study Location: The research was conducted at the private residence on Jalan Nurul Ihsan I, Jagakarsa, South Jakarta, Indonesia.

Study Duration: Oktober 2022 to Januari 2023.

Collection of Matrix Protein p17 Sequencing Data: The virus sequence data was taken from the PDB website: (https://www.rcsb.org/). The sequencing data collection aimed to determine the T and B cells to be studied.

Antigen, Allergy, and Toxicity Analysis: The obtained sequence was then subjected to antigen, allergy, and toxicity tests. These tests aimed to select sequences that met the criteria for vaccine development. For the antigen test, it could be performed on the website: (http://www.ddg-pharmfac.net/vaxijen/VaxiJen/VaxiJen.html) with target organism: Virus and Threshold: 0.4. Meanwhile, for the allergy test, it was performed on the website: (https://www.ddgpharmfac.net/AllerTOP/). The toxicity test was carried out on the website: (http://webs.iiitd.edu.in/raghava/toxinpred/design.php).

MHC I Epitope Analysis: Analysis of MHC I epitope was carried out on the website: (http://ledb.org) by analyzing the selected non-allergic sequence. By entering the allele code HLA-A01:01 and amino acid length of 9.

MHC II Epitope Analysis: Analysis of MHC II epitope was carried out on the website: (http://iedb.org) by entering the selected sequence with DRB101:02 allele code that was an epitope for Asian races with amino acid length of 15, and percentile rank value less than 10.

B Cell Epitope Analysis: B cell epitope analysis was performed on the website: (http://ledb.org). B cell analysis was carried out by entering the selected sequence on the IEDB website, with a peptide length of 10-20.

Protein Homolog Analysis: The selected epitopes were subjected to homology testing on the website: (http://blast.ncbi.nlm.nih.gov) to determine the amino acid residues with body surface receptor.

Design With Adjuvant and Linker: The method used to design vaccine candidates was design with adjuvant and linker using adjuvants as a prefix. By entering the formula as follows: Adjuvant-EAAAK-(MHC I)-AAY-(MHC I)-GPGPG-(MHC II)-KK-(B Cell)-KK-(B Cell). The selected adjuvant was based on the literature used.

Validation of Vaccine Candidates:

Physicochemical Analysis of Vaccine Candidates: Physicochemical tests were carried out on the website: (https://web.expasy.org/protparam/). This analysis was carried out to determine the physicochemical properties of vaccine candidates.

Prediction of Secondary Structure: The secondary structure prediction aimed to see the protein structure. The analysis was carried out on the website: (http://bioinf.cs.ucl.ac.uk/psipred).

Visualization of 3D Vaccine Candidate: Enter the results of the vaccine candidate design on the website: (http://galaxy.seoklab.org/). Visualization is done on the galaxy TBM menu. After obtaining the visualization results on the galaxy TBM, further visualization refinement is carried out on the galaxy refine menu to improve the 3D visualization of the vaccine candidate from the TBM template.

Molecular Docking: Molecular docking was carried out on the Haddock web server on the website: (https://wenmr. science.uu.nl/haddock2.4/) using the TLR4 ligand with PDB ID: 4G8A, which was saved in PDB format, and then docked with the visualization results of the vaccine candidate.

Ligplot and Yasara Analysis: The docking results were then analyzed using LigPlot and Yasara applications to see the visualization of the docking results.

RESEARCH RESULT

Collection of Matrix Protein p17 Sequencing Data

The search for sequence data from the matrix protein p17 was conducted on the Protein Data Bank web server. Based on the search results, the sequence of the matrix protein p17 with the PDB code 2HMX was obtained. In selecting and taking the sequence, the criteria were that the sequence had to be antigenic, non-allergic, and non-toxic.

Antigen, Allergy, and Toxicity Analysis

The sequence used for this research was chosen by determining sequences that met the criteria including being antigenic, non-allergenic, and non-toxic in accordance with the regulations, as presented in Table no 1.

Table 1. The antigen, non-allergic, and solubility evaluation of the selected sequences was assessed.

Proteins	Sequence	Antigen	Allergen	Toxic
Matrix Protein p17	HMGARASVLSG GELDKWEKIRL RPGGKKQYKLK HIVWASRELERF AVNPGLLETSE GCRQILGQLQPS LQTGSEELRSLY NTIAVLYCVHQ RIDVKDTKEAL DKIEEEQNKSK KKAQQAAADT GNNSQVSQNY	0,5413	Non Allergen	Non Toxic
Webserver	PDB	VaxiJen V.2.0	AllerTOP V.2.0	Toxsinpred

MHC I Epitope Analysis

Analyzed MHC I epitopes using HLA-A*01-01 allele with a 9-allele length to facilitate determination of the recognized epitopes, and the selected percentile rank value was less than 1. Epitopes with a percentile rank value less than 1 could easily be recognized by antibodies. MHC I epitope analysis was performed on the IEDB webserver. The results of the MHC I epitope analysis can be seen in table no 2.

Table 2. MHC I Epitope Analysis

T Cell	Allele	Start	End	Length	Epitope	Percentile rank
MHC I	HLA-A*01-01	72	80	9	GSEEL RSLY	0.02
	HLA-A*01-01	125	133	9	NNSQV SQNY	0.53

MHC II Epitope Analysis

MHC II epitope analysis was performed using the HLA DRB1*01-02 allele with a 15-allele length, and the selected percentile rank value was less than 10. Epitopes with a percentile rank value less than 1 could easily be recognized by antibodies. MHC II epitope analysis was performed on the IEDB webserver. The results of the MHC II epitope analysis can be seen in table no 3.

Table 3. MHC II Epitope Analysis

T Cell	Allele	Start	End	Length	Epitope
MHC II	HLA-DRB1*01-	74	88	15	EELRSLYNTIAVLY

02				
HLA-DRB1*01-02	75	89	15	ELRSLYNTIAVLYCV
HLA-DRB1*01-02	72	86	15	GSEELRSLYNTIAVL
HLA-DRB1*01-02	76	90	15	LRSLYNTIAVLYCVH
HLA-DRB1*01-02	70	84	15	QTGSEELRSLYNTIA
HLA-DRB1*01-02	73	87	15	SEELRSLYNTIAVLY
HLA-DRB1*01-02	71	85	15	TGSEELRSLYNTIAV

B Cell Epitope Analysis

In the B cell analysis performed on the IEDB webserver using the p17 matrix protein sequence, the results obtained are presented in table no 4.

Table 4. B Cell Epitope Analysis

No.	***Start***	***End***	**Epitop**	***Length***
1.	41	52	ELERFAVNPGLL	12
2.	66	78	QPSLQTGSEELRS	13

B cell analysis of the selected epitopes was performed using the BepiPred method. The accuracy of the BepiPred method for predicting B cell epitopes reaches 80%. The BepiPred method predicts the location of linear B cell epitopes.

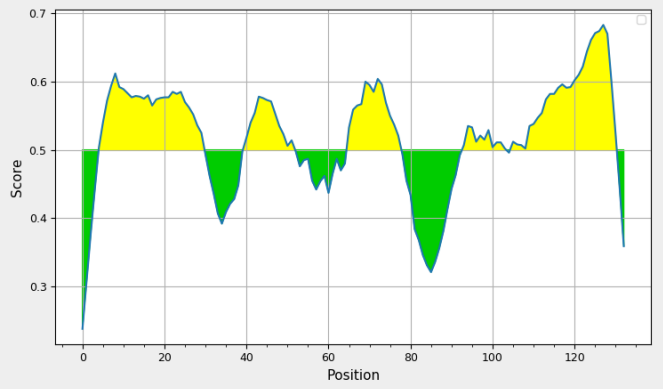

Figure no 1 : Prediction result of B cell epitop

Epitop Analysis

The selected epitope underwent homolog analysis to determine the selected epitope as a vaccine candidate that is not harmful to the body. The results of the homolog analysis can be seen in Table no 5.

Table 5. Epitop Analysis

Lymphocytes	Epitope	Antigenicity	Allergenicity	Toxicity	Human Homology
MHC I	**GSEELRSLY***	Antigen	Non-allergen	Non-toxic	Non-homologous
	NNSQVSQNY*	Antigen	Non-allergen	Non-toxic	Non-homologous
MHC II	EELRSLYNTIAVLY	Non-antigen	Allergen	Non-toxic	Non-homologous
	ELRSLYNTIAVLYCV	Antigen	Allergen	Non-toxic	Non-homologous
	GSEELRSLYNTIAVL	Antigen	Allergen	Non-toxic	Non-homologous
	LRSLYNTIAVLYCVH	Non-antigen	Allergen	Non-toxic	Non-homologous
	QTGSEELRSLYNTIA	Non-antigen	Non-allergen	Non-toxic	Non-homologous
	SEELRSLYNTIAVLY	Non-antigen	Allergen	Non-toxic	Non-homologous
	TGSEELRSLYNTIAV*	Antigen	Non-allergen	Non-toxic	Non-homologous
Sel B	ELERFAVNPGLL	Non-antigen	Non-allergen	Non-toxic	Non-homologous
	QPSLQTGSEELRS*	Antigen	Non-allergen	Non-toxic	Non-homologous
	DVKDTKEALDK*	Antigen	Non-allergen	Non-toxic	Non-homologous

Design With Adjuvant and Linker

After obtaining the selected epitope from the antigen analysis, which is non-allergic, non-toxic, and non-homologous, the vaccine candidate design was carried out. The vaccine candidate design was done by incorporating an adjuvant into the selected epitope sequence, which would be linked with an appropriate linker. Adjuvants are protein vaccines that can enhance the immunogenicity of the vaccine.

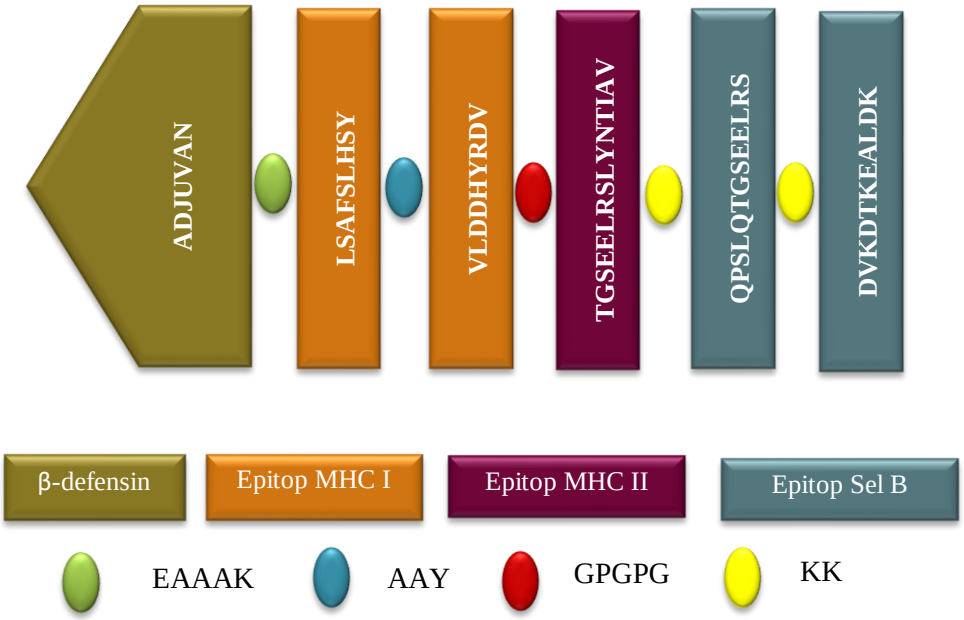

Figure 2. Amino Acid Sequence of HIV Vaccine Candidates

Validation of Vaccine Candidates:

Physicochemical Analysis of Vaccine Candidates

Physicochemical analysis was carried out to determine the physical and chemical properties of the designed vaccine candidate. The results of the physicochemical analysis of the vaccine candidate can be seen in Table no 6.

Table 6. Physicochemical Analysis

Characteristics	**Physicochemical**	**Website**
Number of amino acids	116	ProtParam
Molecular weight	12871.66 g/mol	ProtParam
Theoretical pl	9.58	ProtParam
No. Negatively charged residues (Asp+Glu)	13	ProtParam
No.Positively charged residues (Arg+Lys)	24	ProtParam
Extinction coefficient	7825 m^2/mol	ProtParam
Instability index	59.77	ProtParam
Aliphatic index	64.74	ProtParam
Grand average of hydropathicity (GRAVY)	-0.958	ProtParam

Prediction of Secondary Structure

The prediction of the secondary structure that has been performed on the PSIPRED webserver indicates the formation of helix, strand, and coil structures caused by interactions between C, O, and NH on amino acids in the polypeptide chain.

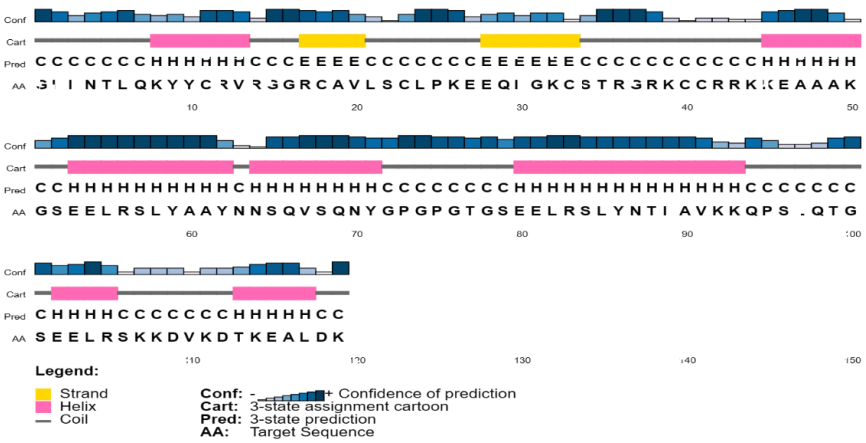

Figure 3. Secondary structure prediction of constructed HIV-1 vaccine protein using PSIPRED server.

Visualization of 3D Vaccine Candidate

The 3D visualization test of the vaccine candidate was carried out on the Galaxy Seoklab webserver. The vaccine candidate visualization method was carried out twice, first by visualizing the vaccine candidate using Galaxy TBM and then by visualizing the vaccine candidate using Galaxy Refine.

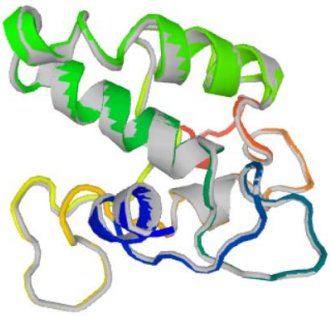

Figure 4. 3D Visualization of HIV-1 Vaccine Candidates

Molecular Docking

Molecular docking was performed using the Haddock webserver, with the aim of predicting the binding between the vaccine candidate and the receptor. The docking score results can be seen in Table no 7.

Table 7. Docking score results

Model	Score
Cluster 1	-89.7
Cluster 7	-69.6
Cluster 3	-66.9
Cluster 8	-64.1

Cluster 6	-61.5

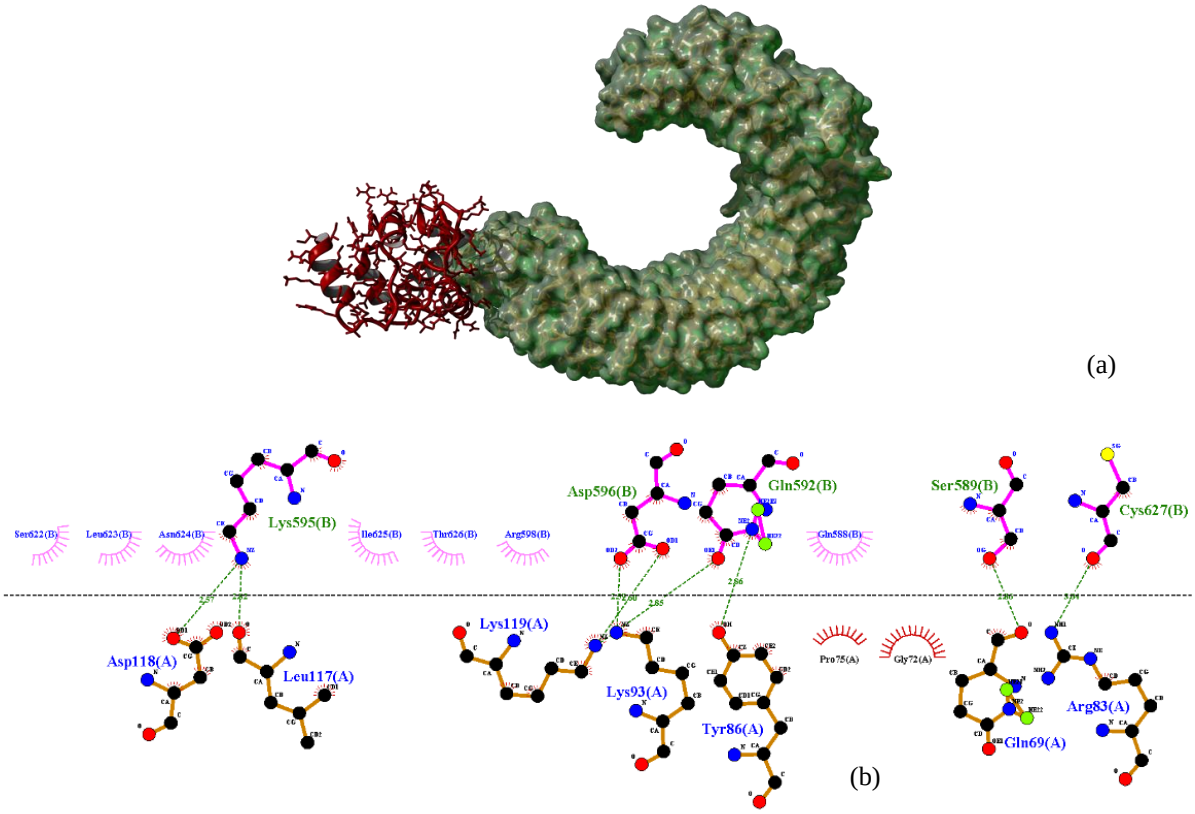

Figure no 5 : Visualization of HIV-1 Vaccine Candidates. (a) 3D visualization
(b) 2D visualization

DISCUSSION

The HIV-1 p17 matrix protein was a structural protein located inside the virus membrane that played a key role throughout the HIV-1 life cycle. The p17 matrix protein can be detected in blood at nanomolar concentrations. After secretion, p17 accumulates and persists in various organs and tissues. In this study, the p17 matrix protein was used as the protein from the HIV virus to design an epitope-based vaccine, where the sequence from the p17 matrix protein was studied.

The sequence used was chosen by determining sequences that met criteria including being antigenic, non-allergic, and non-toxic. The sequence used was antigenic in order to stimulate the immune system to prevent virus infection. Using a threshold value of 0.4, this determination aimed to see the accuracy and sensitivity of the test.

Antigens can include whole or parts of microbes, chemical components of bacterial structures, non-microbial chemical components (egg whites, pollen, tissue or organ transplants). A certain small part of a large antigen molecule can trigger an immune response called an epitope or antigenic determinant.

Epitopes can belong to foreign proteins and self-proteins, and they can be categorized as conformational or linear, depending on their structure and integration with the paratope. T cell epitopes are presented on the antigen-presenting cell (APC) surface, where they bind to major histocompatibility complex (MHC) molecules to induce an immune response.

The general concept behind peptide vaccines is based on a chemical approach to synthesize T cell and B cell epitopes that are identified as immunodominant and capable of inducing specific immunity. Targeting the molecular structure of B cell epitopes can be conjugated with T cell epitopes to form immunogenicity. T cell epitopes bind to MHC to induce an immune response.

The Major Histocompatibility Complex (MHC) protein is divided into two classes, MHC class I and MHC class II. MHC class I and class II play a critical role in the adaptive immune system branch. Both protein classes share the task of presenting surface peptides for T cell recognition. The MHC I peptide complex is presented on nucleated cells and recognized by cytotoxic CD8 T cells. Presentation from MHC II may activate CD4 T cells, leading to coordination and effector cell regulation. T cell epitope analysis aims to identify the shortest peptide in the antigen that is capable of inducing T cell. In the MHC I analysis, there were two epitopes estimated to be HIV vaccine candidates with a percentile rank value of less than one, namely GSEELRSLY and NNSQVSQNY. In the MHC II analysis results, there were seven epitopes estimated to be HIV-1 vaccine candidates with a percentile rank value of less than ten, namely EELRSLYNTIAVLY, ELRSLYNTIAVLYCV, GSEELRSLYNTIAVL, LRSLYNTIAVLYCVH, QTGSEELRSLYNTIA, SEELRSLYNTIAVLY, TGSEELRSLYNTIAV.

For B cell activation, it is mediated by the antigen-IgM binding to the membrane-bound IgM antigen on the surface of the B cell. It is then stated that after antigen-IgM is activated, the B cell will differentiate into plasma cells and secrete antibodies, either in the form of secreted IgM, IgG, or IgA. All the antibody classes produced (IgM, IgG, or IgA) will also recognize the same epitope that induces B cell activation. The B cell analysis results found three epitopes estimated to be vaccine candidates with peptide lengths of 10-20, namely ELERFAVNPGLL, QPSLQTGSEELRS, and DVKDTKEALDK. The selection of epitopes with amino acid lengths of 10-20 is an epitope with an appropriate molecular weight and does not pose excess molecular weight if made into a vaccine candidate. With excessive molecular weight, the epitope cannot be used as a vaccine candidate.

Selected epitopes from the analysis results that meet the criteria as an antigenic epitope, non-allergenic, non-toxic, and non-homologous were designed by combining an adjuvant linked with a linker, as follows: adjuvant-EAAAK-GSEELRSLY-AAY-NNSQVSQNY-GPGPG-TGSEELRSLYNTIAV-KK-QPSLQTGSEELRS-KK-DVKDTKEALDK. A vaccine containing β-defensin as an adjuvant is capable of activating the main innate antiviral immune response and mediating other immunomodulatory activities against a range of viruses. This is what makes the adjuvant effective when conjugated with an antigen.

Furthermore, the adjuvant is combined with an EAAAK linker to enhance the antigenicity and immunogenicity of the peptide-based vaccine. Followed by an MHC I epitope connected with the AAY linker. The AAY linker (Ala-Ala-Tyr) is a protease cleavage site used to increase epitope presentation and enhance protein stability. The linker between MHC I and MHC II is GPGPG, known as the glycine-proline linker to prevent epitope junctional. For the linker between MHC II and B cell is KK. The bi-lysine linker is a linker that establishes immunogenic activity of a vaccine.

The HIV-1 vaccine candidate was analyzed to determine its physicochemical properties, where the results showed that the vaccine candidate contained 116 amino acids, with a molecular weight of 12871.66 g/mol, an isoelectric point (pI) value of 9.58. The total negatively charged residues (Asp+Glu) were 13 residues and the positively charged residues (Arg+Lys) were 24 residues. The molar absorption coefficient or commonly called the extinction coefficient in the vaccine candidate test resulted in a length of 7825 m2/mol. Furthermore, the stability index value of the vaccine candidate was 59.77, where the stability index is a measure of protein that can estimate the stability of a protein. Proteins with a stability index value less than 40 are predicted to be stable, while those larger than 40 are predicted to be unstable. The aliphatic index has a value of 64.74. The GRAVY value of the vaccine candidate was -0.958, indicating that the target protein is hydrophilic.

Next, to predict the binding between the vaccine candidate and the surface receptor of the body, molecular docking was carried out, where the best docking score resulted in the lowest score value. Molecular docking between the protein and the receptor resulted in a lowest energy score of -89.7. A negative sign on the energy value that gets smaller indicates a strong complex formed between the ligand and the standard. This is due to an increase in torsional energy from the complex, making the enzyme and ligand complex stable.

CONCLUSIONS AND RECOMMENDATIONS

Based on the research results, the peptide sequence that had the potential as an HIV-1 vaccine candidate was as follows, adjuvant-EAAAK-GSEELRSLY-AAY-NNSQVSQNY-GPGPG-TGSEELRSLYNTIAV-KK-QPSLQTGSEELRS-KK-DVK DTKEALDK. The physicochemical properties of the HIV vaccine candidate had 116 amino acid residues, a molecular weight of 12871.66 g/mol, a theoretical pI of 9.58, 13 negatively charged residues, 24 positively charged residues, an extinction coefficient of 7825 m2/mol, a stability index of 59.77, an aliphatic index of 64.74, and an average hydrophobicity of -0.958. The vaccine design using the p17 protein matrix was not homologous or did not cause autoimmune responses when used as a vaccine candidate.

ADVANCED RESEARCH

Further research was expected to be conducted on the design of an HIV vaccine using other proteins such as the p24 protein present in the HIV virus, as

the matrix protein p17 was found to be less stable when considered as a vaccine candidate.

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