

Prediction of Epitope based Peptides for Vaccine Development from Complete Proteome of Novel Corona Virus (SARS-COV-2) Using Immunoinformatics

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Abstract

COVID-19 is an infectious disease caused by a newly discovered corona virus SARS-COV-2. It is the most dangerous epidemic existing currently all over the world. To date, there is no licensed vaccine and not any particular efficient therapeutic agent available to prevent or cure the disease. So development of an effective vaccine is the urgent need of the time. The proposed study aims to identify potential vaccine candidates by screening the complete proteome of SARS-COV-2 using the computational approach. From 14 protein entries in UniProtKB, 4 proteins were screened for epitope prediction based on consensus antigenicity predictions and various physico-chemical criteria like transmembrane domain, allergenicity, GRAVY value, toxicity, stability index. Comprehensive analysis of these 4 antigens revealed that spike protein (P0DTC2) and nucleoprotein (P0DTC9) show the greatest potential for experimental immunogenicity analysis. These 2 proteins have several potential CD4+ and CD8+ T-cell epitopes, as well as high probability of B-cell epitope regions as compared to well-characterized antigen the matrix protein 1 [Influenza A virus (H5N1)]. In addition, the epitope SIIAYTMSL predicted from spike protein (P0DTC2) and epitope SPRWYFYYL predicted from nucleoprotein (P0DTC9) exhibited more than 60% population coverage in the target populations Europe, North America, South Asia, Northeast Asia taken in this study. These epitopes have also been found to exhibit highly significant TCR–pMHC interactions having a joint Z value of 4.51 and 4.37 respectively. Therefore, this analysis suggests that the predicted epitopes might be suitable vaccine candidates and should be subjected to further in-vivo and in-vitro studies.

Keywords SARS-COV-2 · Covid-19 · Vaccine · Epitope · MHC

Introduction

COVID-19 is a deadly disease caused by SARS corona viruses world-wide. More than 59 million (59,481,31) confirmed cases and more than 1 million (1,404,542) deaths have been reported to WHO till 25 November 2020. A pneumonia of unknown cause detected in Wuhan, China was first reported to the WHO Country Office in China on 31 December 2019. The outbreak was declared a Public Health

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Emergency of International Concern on 30 January 2020. On 11 February 2020, WHO announced a name for the new coronavirus disease: COVID-19.

SARS-COV-2 has round or elliptic and often pleomorphic form, and a diameter of approximately 60–140 nm (Cascella et al. 2020). It is a positive sense ssRNA virus of about 30 kb genome size. This virus belongs to family coronaviridae and genus Betacoronavirus. SARS-COV-2 genome contains two flanking untranslated regions (UTRs) and a single long open reading frame encoding a polyprotein. The 2019nCoV genome is arranged in the order of 5'-replicase (orf1/ ab)-structural proteins [Spike (S)-Envelope (E)-Membrane (M)-Nucleocapsid (N)]-3' (Chan et al. 2020). Two-thirds of viral RNA, mainly located in the first ORF (ORF1a/b) translates two polyproteins, pp1a and pp1ab, and encodes 16 non-structural proteins (NSP), while the remaining ORFs encode accessory and structural proteins. The rest part of virus genome encodes four essential structural proteins, including spike (S) glycoprotein, small envelope (E) protein, matrix (M) protein, and nucleocapsid (N) protein (Cui et al. 2019), and also several accessory proteins, that interfere with the host innate immune response.

Based on virus genome sequencing results and evolutionary analysis, bat has been suspected as natural host of virus origin, and SARS-COV-2 might be transmitted from bats via unknown intermediate hosts to infect humans. Direct contact with intermediate host animals or consumption of wild animals was suspected to be the main route of SARS-COV-2 transmission. However, the source(s) and transmission routine(s) of SARS-COV-2 remain elusive (Guo et al. 2020).

COVID-19 affects different people in different ways. Symptoms may appear 2–14 days after exposure. Serious symptoms include difficulty in breathing, chest pain and loss of speech or movement. The most common symptoms of COVID-19 are fever, dry cough, and tiredness. Other symptoms that are less common and may affect some patients include aches and pains, nasal congestion, headache, conjunctivitis, sore throat, diarrhoea, loss of taste or smell or a rash on skin or discoloration of fingers or toes.

Transmission of the disease occurs mainly through person to person. When the person infected with COVID-19 coughs, sneezes or speaks, small droplets expelled from them land on surfaces and objects around them. Other people then catch COVID-19 by touching these objects or surfaces, then touching their eyes, nose and mouth or by breathing these droplets. Major complications due to COVID-19 include acute respiratory failure, pneumonia, acute respiratory distress syndrome, acute kidney injury, acute liver injury, acute cardiac injury, septic shock, blood clots, rhabdomyolysis, disseminated intravascular coagulation, secondary infections (Zaim et al. 2020).

Researchers worldwide are working around the clock to find a vaccine against SARS-CoV-2, the virus causing the COVID-19 pandemic. There are no effective vaccines or specific antiviral drugs for COVID-19 (Dhama et al. 2020). Possible vaccines and some specific drug treatments are under investigation. Three vaccines, two adenoviral vector vaccines and a protein-based vaccine, have been given early or limited approval without waiting for the results of phase III trials. Sputnik V formerly known as Gam-COVID-Vac developed by the Gamaleya Research Institute in Moscow, Russia, was approved by the Ministry of Health of the Russian Federation on 11 August 2020. Another vaccine developed by the Chinese company CanSino Biologics, was approved by the Chinese military in June 2020 for a year as "a specially needed drug". A second vaccine in Russia, EpiVacCorona, developed by the State Research Center of Virology and Biotechnology, has also been granted regulatory approval On 14 October 2020, also without entering Phase 3 clinical trials (Robinson 2020 online). According to WHO Draft landscape of COVID-19 vaccine candidates 12 November 2020, there are 48 vaccine candidates in clinical evaluation and 164 in preclinical evaluation.

The conventional approach to vaccine development is based on dissection of the pathogen using biochemical, immunological and microbiological methods. Although successful in several cases, this approach has several limitations. This method can employ many years to identify a protective and useful antigen, and has failed to provide a vaccine against those pathogens that did not have obvious immunodominant protective antigens. The availability of complete genome sequences in combination with novel advanced technologies, such as bioinformatics, microarrays and proteomics, have revolutionized the approach to vaccine development and provided a new impulse to microbial research (Capecchi et al. 2004). To use computers to rationally design vaccines starting with information present in the genome, without the need to grow the specific microbe, this new approach was denominated 'reverse vaccinology' (Rappuoli 2000). The first example of reverse vaccinology approach is the development of a vaccine against serogroup B Neisseria minigitidis (MenB), a pathogen that causes 50% of the meningococcal meningitis worldwide. It took less than 18 months to identify more and some novel vaccine candidates in MenB than had been discovered during the past 40 years by conventional methods (Pizza et al. 2000). Reverse vaccinology is now being applied to many bacterial, viral and eukaryotic pathogens and has been successful in all cases in providing novel antigens for the design of new vaccines (Bagnoli et al. 2011). Vaccine candidates identified from a pathogen's genome or proteome can then be expressed as recombinant proteins and tested in appropriate in vitro or in vivo models to assess immunogenicity and protection (Seib et al. 2000).

In the present study, SARS-COV-2 (NC_045512.2) reference strain, which is known to cause COVID-19 pandemic was undertaken to characterize its antigens as potential vaccine candidates.

Materials and Method

Retrieval of Proteome Data Set

The complete proteome sequence of SARS-COV-2 has been retrieved from Viralzone Expasy server (viralzone.expasy. org). The sequences have been stored as fasta file containing all 14 annotated UniProtKB protein entries. A well characterized viral antigen showing proper immune response in humans the matrix protein 1 [Influenza A virus (H5N1)] has been taken as control to compare and validate the outcomes. It has been tested as an adjuvanted virosomal H5N1

vaccine and found to induce a balanced Th1/Th2 CD4(+) T cell response in man (Pederson et al. 2014).

Antigenicity Prediction

Antigenicity prediction of all the protein sequences has been performed to determine their overall possible role in initiating an immune response. Consensus antigenicity predictions have been performed using Vaxijen and ANTIGENpro tools. VaxiJen is the first server for alignment-independent prediction of protective antigens. It was developed to allow antigen classification solely based on the physicochemical properties of proteins without recourse to sequence alignment. It is freely available through https://www.ddg-pharmfac.net/ vaxijen/VaxiJen/VaxiJen.html (Doytchinova and Flower 2007). ANTIGENpro is a sequence-based, alignment-free and pathogen-independent predictor of protein antigenicity. The predictions are made by a two-stage architecture based on multiple representations of the primary sequence and five machine learning algorithms. ANTIGENpro is integrated in the SCRATCH suite of predictors available at: http://scratch. proteomics.ics.uci.edu (Magnan et al. 2010).

Characterization of Predicted Antigenic Proteins

Genome-wide characterization of vaccine candidates has been performed using various computational tools. Transmembrane regions have been predicted using TMHMM web server. It is based on hidden Markov model (Krogh et al. 2001). Assessment of allergenic potential has been carried out using AllerCatPro tool. It is entropy-adjusted hexamer hit approach as well as switching from a linear sequence window similarity to a B-cell epitope-like 3D surface similarity with predicted structures for 74% of all known allergens in a workflow guided by safety rationale (Maurer et al. 2019). Physical chemical parameters are calculated using ProtParam tool available at expasy. These parameters include the molecular weight, theoretical pI, instability index, aliphatic index and grand average of hydropathicity (GRAVY) (Gasteiger et al. 2005).

B Cell Epitope Prediction

The antigenic regions of protein recognized by the binding sites of immunoglobulin molecules are called B cell epitopes (Van Regenmortel 1993). B cell epitopes can be classified into two categories: conformational/ discontinuous epitope, where residues are distantly separated in the sequence and brought into physical proximity by protein folding and linear/continuous epitope comprised of a single continuous stretch of amino acids within a protein sequence that can react with anti-protein antibodies (Barlow et al. 1986). The designing of conformational epitopes is difficult and so experimental B cell epitopes largely include linear epitopes. A web server, BepiPred has been used to determine the probability of presence of linear B cell epitopes in the selected antigen sequences. It is based on a random forest algorithm trained on epitopes annotated from antibody-antigen protein structures. It is available at http://www.cbs.dtu.dk/services/ BepiPred/ (Jespersen et al. 2017).

T Cell Epitope Prediction

T-cell epitope prediction aims to identify the shortest peptides within an antigen that are able to stimulate either CD4 or CD8 T-cells (Ahmed and Maeurer 2009). T-cell epitopes are presented on the surface of an antigen presenting cell (APC), where they are bound to major histocompatibility (MHC) molecules in order to induce immune response (Madden 1995). Cytotoxic T lymphocytes (CTL) epitope prediction has been performed using NetCTL, a web based tool designed for predicting human CTL epitopes in any given protein. It does so by integrating predictions of peptide MHC class I binding, proteasomal C terminal cleavage and TAP transport efficiency. MHC class I binding and proteasomal cleavage is performed using artificial neural networks. TAP transport efficiency is predicted using weight matrix. Peptides with a combined prediction score greater than or equal to default threshold value (0.75) are marked as potential HLA class I supertype CTL epitopes. NetCTL provides a comprehensive prediction about epitopes binding to 12 HLA class I supertypes including 5 HLA-A [A1, A2, A3, A24, A26] and 7 HLA-B [B7, B8, B27, B39, B44, B58, B62] (Larsen et al. 2007). It is available at http://www.cbs. dtu.dk/services/NetCTL. These predicted CTL epitopes have been again subjected to antigenicity prediction using Vaxijen server to assure the credibility. Furthermore, to predict binding of peptides to HLA-DR, MHC class II alleles, NetMH-CII 2.2 server has been used. Predictions can be obtained for 25 HLA-Dr alleles, 20 HLA-DQ, 9 HLA-DP, and 7 mouse H2 class II alleles. It is based on artificial neural networks and publicly available at www.cbs.dtu.dk/services/NetMH CII (Nielsen and Lund 2009).

Population Coverage Analysis

T cells recognize a complex between a specific major histocompatibility complex (MHC) molecule and a particular pathogen-derived epitope. A given epitope will elicit a response only in individuals that express an MHC molecule capable of binding that particular epitope. MHC molecules are extremely polymorphic and over a thousand different human MHC (HLA) alleles are known (Bui et al. 2006). Specific HLA alleles are expressed at dramatically different frequencies in different ethnicities (Gjertson and Lee 1998; Imanishi et al. 1992). A web based tool, IEDB population coverage, has been used for population coverage analysis. This method calculates the fraction of individuals predicted to respond to a given epitope or epitope set on the basis of HLA genotypic frequencies and on the basis of MHC binding and/or T cell restriction data (Bui et al. 2006). It can be accessed through http://tools.iedb.org/population/. COVID-19 has affected all over the world, in this study Europe, North America, South Asia, Northeast Asia have been taken as target populations. The analysis focused on MHC I because of the fact that viral peptides are presented only on MHC I via the endogenous pathway (Srivastava et al. 2016).

pMHC-TCR Interaction Analysis

Proper interaction of peptide-MHC complex with TCR is very important for adaptive immune responses. PAComplex server has been utilized for this purpose. The PAComplex is a web server for predicting TCR-pMHC interactions and inferring antigen families across organisms, of a query protein or a set of peptides. This server first identifies significantly similar TCR-pMHC templates (joint Z-value ≥ 4.0) of the query by using antibody-antigen and protein-protein interacting scoring matrices for peptide-TCR and pMHC interfaces, respectively (Liu et al. 2011). The joint Z-value (Jz) is defined as:

 $\sqrt{Jz} = Z MHC \times Z TCR$ (Marrack et al. 2008)

Here, J $z \ge 4.0$ is considered a significant similarity according to the statistical analysis.

PAComplex then identifies the homologous peptide antigens of these hit templates from complete pathogen genome databases and experimental peptide databases. Finally, the server outputs peptide antigens and homologous peptide antigens of the query and displays detailed interacting models of hit TCR-pMHC templates (Liu et al. 2011). The PAComplex server is available at http://PAcomplex.life.nctu. edu.tw. Here, the CTL epitope set predicted by NetCTL and optimized by IEDB for the different target population, has been used as the target peptide set and TCR-pMHC interactions have been analyzed.

Results and Discussion

Selection of Antigens

The complete protein repertoire of SARS-COV-2 has been screened for proteins having sufficient antigenicity property. Consensus predictions have been made using Vaxijen and ANTIGENpro tools at pre-defined threshold value 0.4 for both. Out of 14 proteins, 7 have shown antigenic probability \geq 0.4. Therefore, based on consensus prediction these 7 antigenic proteins have been taken for further analysis (Table 1). Control antigen has been found to be antigenic by both the tools.

Characterization of Selected Antigens

Proteins with more than one transmembrane (TM) region have been found to be difficult to clone, express and purify; thus 7 antigenic proteins predicted in the previous step have been subjected to predict presence of transmembrane domains using TMHMM server. Out of 7 antigenic proteins, 2 antigens (P0DTC1, P0DTD1) have been predicted to contain 14 TM regions, 1 antigen (PODTC3) with 3 TM regions, 2 antigens (P0DTC2, P0DTC7) with 1 TM region and 2 antigens (P0DTC9, P0DTD2) with no TM regions (Table 1). So, these 4 antigens (P0DTC2, P0DTC7, P0DTC9, P0DTD2) are taken for further analysis. The control antigen has also not shown any TM regions. In allergenicity prediction using AllerCatPro tool, all the 4 antigenic proteins have been predicted as non- allergen. The control antigen has also been found to be non-allergen. The physical chemical parameters calculated using ProtParam tool has been shown in Table 2.

Table 1 List of proteins predicted to be antigenic with corresponding antigenic probabilities

Protein no	UniProtKB id	Protein name	Antigenic Probability		No. of TM regions	
			VaxiJen	ANTIGENpro	predicted using TMHMM	
1	P0DTC1	Replicase polyprotein 1a (pp1a)	0.47	0.64	14	
2	P0DTD1	Replicase polyprotein 1ab (pp1ab)	0.46	0.68	14	
3	P0DTC2	Spike glycoprotein (S)	0.46	0.71	1	
4	P0DTC3	ORF3a protein (NS3a)	0.49	0.40	3	
5	P0DTC7	ORF7a protein	0.64	0.40	1	
6	P0DTC9	Nucleoprotein (N)	0.50	0.93	0	
7	P0DTD2	ORF9b protein	0.90	0.74	0	
Control	Q9Q0L8	Matrix protein 1	0.47	0.86	0	

UniprotKB id	Protein name	Molecular weight (KDa)	Theoretical pI	Instability Index	Aliphatic Index	GRAVY
P0DTC2	Spike glycoprotein (S)	141.17	6.24	33.01	84.67	- 0.079
P0DTC7	ORF7a protein	13.74	8.23	48.66	100.74	0.318
P0DTC9	Nucleoprotein (N)	45.62	10.07	55.09	52.53	- 0.971
P0DTD2	ORF9b protein	10.79	6.56	33.11	105.46	- 0.085
Q9Q0L8 (Control)	Matrix protein 1	27.85	9.42	38.72	82.90	- 0.246

 Table 2
 Physical chemical parameters calculated using ProtParam tool

Antigen P0DTC7 has been shown instability index > 40 i.e. 48.66, GRAVY value positive i.e. 0.318 so it has been removed from further analysis. Thus, based on screening so far, finally 3 candidate antigens (P0DTC2, P0DTC9, P0DTD2) have been selected for epitope prediction.

B Cell Epitope Prediction

According to BepiPred linear B cell epitope predictions at threshold 0.45, high probability of B cell epitope has been found in all the three antigens. Antigens P0DTC2, P0DTC9 and P0DTD2 have been predicted to have 25, 9 and 2 regions respectively as probable B cell epitopes regions. Similar criteria set has been used for control antigen and 6 regions have been predicted as probable B cell epitope regions.

T Cell Epitope Prediction

For HLA class I supertypes, based on highest value of combined score obtained using NetCTL, a total of 419 putative CTL epitopes have been predicted for antigen P0DTC2, 104 putative CTL epitopes have been predicted for antigen P0DTC9 and 33 putative CTL epitopes have been predicted for antigen P0DTD2. The control antigen has been predicted to show 88 putative CTL epitopes. Antigenicity analysis of these predicted CTL epitopes using Vaxijen server at threshold 0.4 has shown that many of them have been found to be non-antigen. So those non-antigenic peptides have been removed and peptides predicted to bind more than one HLA class I supertype have been selected. Thus, 53 putative CTL epitopes have been selected from antigen P0DTC2, 10 putative CTL epitopes have been selected from antigen P0DTC9 and 8 putative CTL epitopes have been selected from antigen P0DTD2 for further analysis as listed in Table 3.

For HLA class II supertypes using NetMHC II algorithm, 341, 79 and 33 putative HTL epitopes have been predicted for P0DTC2, P0DTC9 and P0DTD2 respectively. The control antigen has been predicted 67 HTL epitopes binding to 15 HLA-DR supertypes.

Population Coverage Analysis

Epitope vaccines trigger an immune response by confronting the immune system with immunogenic peptides. Binding of these peptides to proteins from the major histocompatibility complex (MHC) is crucial for immune system activation. However, since the MHC is highly polymorphic, crucial step in design of a peptide vaccine is the selection of the set of epitopes which yields the best immune response in a given population or individual (Jain et al. 2019). It has been demonstrated that a correlation exists between immunogenicity and MHC class I binding affinity (Sette et al. 1994). It is, therefore, reasonable to use MHC class I binding affinity prediction methods for the prediction of immunogenicity.

CTL epitope sets obtained in the previous step have been taken as input for population coverage analysis. IEDB population coverage server outputs percentage population coverage of individual epitope in the epitope set for all the target populations taken. Table 4 shows the top scoring epitopes and their respective population coverage percentage.

pMHC-TCR Interaction Analysis

T cells do not recognize soluble native antigen but rather recognize antigen that has been processed into antigenic peptides, which are presented in combination with MHC molecules. T-cell epitopes must be viewed in terms of their ability to interact with both a T-cell receptor and an MHC molecule. The interaction between the T-cell receptor and an antigen bound to an MHC molecule is central to both humoral and cell-mediated responses (Goldsby et al. 2007). The results obtained in TCR-pMHC interaction analysis using PAComplex are described below.

For peptide set from antigen P0DTC2, the same hit peptide has been obtained for all the four target populations. The epitope SIIAYTMSL has been found to have a joint Z value of 4.51, illustrating that this peptide demonstrates highly

value of 4.37, indicating that this peptide exhibits immensely

valuable pMHC-TCR interactions (Fig. 2). This hit peptide

Table 3 Selected CTL epitopes and their binding to different MHC

Table 3 (continued)

ProteinEpilopeMHC I supertypesPTOTC2 Spike glycoprotein (S)AAUQIPTAMB7, B58AAUQIPTAMB7, B58TLDSKTQSLA2, B39AIVMVTIMLA2, B7TLLALIRRSYA3, B62DEDDSEVU.B39, B44TSNQVAVLYA1, A3, A26, B58, B62EVVKOVKLB7, B8VLROVKLHYA1, B62FAMQMAYRFB58, B62VLFNDGVYA1, B62FANQMAYRFB58, B62VLPENDGVYA1, B62FTISVTTEIA2, A26, B58VLQPKTHILB27, B62FTISVTTEIA2, A26, B58, B62VLQPKTH1.A2, B39, B58FVENVLLPLA2, A26, B58, B62VLQPKTH1.A2, B39, B58GARHVNNSYA1, B58, B62VLQPKTH1.A2, A26, B62GQTGKIADYB58, B62FUTDAVDCALA1, B39, B58KGRYQTSNFB58, B62FUTDAVDCALA1, B39, B58KGRYQTSNFB58, B62SPRPARSAFA24, B58, B62KIADDYNYKLA2, B58GARHVNYYA2, B58, B62KIADYNYKLA2, B58GARHVNYYA2, B58, B62KINSNETPTA2, B58GARHVYYYA1, A3, A26, B58, B62KINSNETPTA2, B58GARHVYYYA1, A3, A58, B62KINSNETPTA2, B58GARHVYYYA1, A3, B58, B62KINSNETPTA2, B58GARHVYYYA1, A3, B58, B62KINSNETPTA2, B58, B62GRASAFY A24, B58, B62KINSNETPTA2, B58, B62GRASAFY A24, B58, B62KINSNETPTA2, B58, B62GRASAFY A26, B58, B62KINSNETPTA2, B58, B62GRASAFY A26, B58	class I supertypes			Protein	Epitope	MHC I supertypes	
PODTC2 AALQIPFAM B7, B58 Spike glycoprotein (S) AI, VMVTIML A2, B7 DEDDSEPVI. B39, B44 TLISKTSGL A2, B39 EPVLKOVKL B7, B5 VLKSVKLIIY A1, A3, A62 FAMGMAYRF B58, B62 VLFPNDOVY A1, A3, B62 FHYNSQPFL B39, B44 VNNONAQAL B7, B62 FLINTYVPA A2, B39 VTPLQEL A24, B39 FRSNLKFF B8, B62 VLFPNDOVY A1, A3, A62 FRSNLKFF B8, B27 WTAGAAAYY A1, A26, B58, B62 FINSVITEI A2, A26, R58 WTPGAGAAL A26, B5 FVENVLIPI A2, A26, R58 WTRGAGAAL A26, B52 GAAAYYVGY A1, B58, B62 YQPYRVVVL A2, A23, B59, B62 GAGICASY B58, B62 YUPAQEKNF A26, B58, B62 IGMINNTVY B58, B62 YUPAQEKNF A24, B58, B58, B62 IGANNYKL A2, B39 SPRWYFY A1, A3, A26, H58, B62 IGANNYKL A2, B39 SPRWYFY A1, A3, A26, H58, B62 IGANNYKL A2, B39 SPRWYFY A1, A3, A26, H58, B62 IGANNYKL A2, B8 SPRWYFY A1, A3, A26, H58, B62 IGANNYKL A2, B8 SPRWYFY A1, A26, B58, B62	Protein	Epitope	MHC I supertypes				
Spike glycopotein (S)CTULLITUREALIONAIVWTIMLA2, B7TULLITURESA3, B62DEDDSEPVLB9, B44TISNQVAVLYA1, A3, A26, B58, B62ENKKRLPFA26, B62VLFGWLHA1, B62FEVX0PFLB3, B44VNQNAQALB7, B62FLNVTYVPAA2, B8VVDPLQPELA24, B39FLNVTYVPAA2, B8VVDPLQPELA24, B39FKSNLKPFB8, B27WTGAGAALA1, A26, B58, B62FVFVLILPLA2, A26, B8, B62VUDPTTLLA2, B39, B58, B62GAAYYVGYA1, B58, B62YUDPYRUVLA2, B39, B58, B62GAAYYVGYA1, B58, B62VUDPKFFLA2, A26, B8, B62GAAYYVGYA1, B58, B62VUNQNCTEVA1, A2, B39, B58, B62GARAYYVGYA1, B58, B62PDITC9DLSPRWYFYA1, A3, A26, B58, B62GARAYYVGYB58, B62Nucleoprotein (N)FREGGGVFHF7, B8IADTNFTIA2, B58CARFARNERA24, B62KIDNYNYKLA2, B39SRSRKIDNYNYKLA2, B39SRSRKINYSKHTPIA2, B8SRSRLALHRSYLA2, B8SRSRKINYSKHTPIA2, B8SRSRLALHRSYLA2, B8SRSRKINYSKHTPIA2, B8SRLALHRSYLA3, A26, B58, B62SRKINYSKHTPIA2, B8SRKINYSKHTPIA2, B8SRLALHRSYLA3, A26, B58, B62SRKINYSKHTPIA2, B8SRKIN	P0DTC2	AALOIPFAM	B7. B58		TFEY VSQPF	A24, B62 A2 B39	
ArMWTINL A2, B7 DEDUSEPVL B39, B44 DEDUSEPVL B39, B44 DEVLKGVKLH7 A1, A3, A26, B58, B62 EVKKGVKL B7, B8 EVK7UPA A2, B8 FFKYSUPF B29, B44 FLWTYVPA A2, B8 FFKSNLKPF B8, B27 FTISVTTEI A2, A26, B58, B62 GAAAYYGY A1, B58, B62 GGGCKHADY B27, B62 GGGCKKADY B58, B62 GGCKKADY B58, B62 GGCKKADY B58, B62 KINYSKHTH A2, B58 KISYDCTMY A1, A3, B58, B62 KINYSKHTH A2, B39 KIADYNYKL A2, B39 KIADYNYKL A2, B39 KIADYNYKL A2, B39 KINSKHTH A2, B8 KISYDCTMY A1, A3, B58, B62 KINYSKHTH A2, B8, B58, B62 KINYSKHTH A2, B8 KISYDCTMY A1, A3, B58, B62 KINYSKHTH A2, B8, B59, B62 KINYSKHTH A2, B8 KISYDCTMY A1, A3, B58, B62 KINYSKHTH A2, B7, B58 LIALLALIRSYL A2, B8 KISYBCTMY B7, B58 LIALLALIRSYL A2, B8 KISYBCTMY A1, B39, B58, B62 KINYSKHTH A2, B7, B58 LIALLALIRSYL A2, B62 KINYBKHTH A2, B7, B58 LIALLALIRSYL A2, B7, B58, B62 KINYSKHTH A2, A2, B7, B58, B62 KINYBKHTH A2, A2, B7, B58, B62 KINYBKHTH A2, A2, B7, B58, B62 KINYBKHTH A2, A2, B7, B58, B62 KINYSKHTH A2, B7, B8, B62 KINYSKHTH A2, A2, B7, B58, B62 KINYBKHTH A2, B7, B8, B62 KINYSKHTH A2, A2, B7, B58, B62 KINYBKHTH A2, B7, B8, B62 K	Spike glycoprotein (S)	C C	.,			A3 B62	
DEDDSERVL.B39, B44ISNQVAULYA1. A3. A26, B58, B62EPVLKGVKLB7, B8VLKGVKLHYA1. A3. B62ESNKKFLPFA26, B58, B62VRFPNITNLB27, B39FEVVSQPFLB39, B44VVNQNQQALB7, B62FLIVTYVPAA2, B8VTDQLQPELA24, B59FRKSNLKPFB8, B7WTGAAAYYA1. A2, B58, B62FISVTTEIA2, A26, B58, B62VLGPRTFLLA2, B58, B62GAAAYYVGYA1, B58, B62VQDVNCTEVA1. A2, B39, B58, B62GQTGKIADYB27, B62VQDVNCTEVA1. A3, A26, B58, B62GQTGKIADYB27, B62VQDVNCTEVA1. A3, A26, B58, B62GQTGKIADYB27, B62VQDVNCTEVA1. A3, A26, B58, B62GQTGKIADYB58, B62VDTC9DLSPRWYTYA1. A3, A26, B58, B62IGAGICASYB58, B62VDTC9DLSPRWYTYA1. A3, A26, B58, B62IDAVDCALA1, B39, B58GA24, B7, B8, 588, B62VFDTU2QFAFSASAFIDAVDCALA1, A3, B58, B62GPAFSASAFA24, B7, B8, B58, B62IDAVDCALA1, A3, B58, B62GPAFSASAFA24, B7, B8, B39, B58KINSWITTIA2, B8SSPDDQIGYA1, A26, B58, B62VTISQVAVLA2, B8SSPDDQIGYA1, A26, B58, B62LPFSNVTWA1, A3, A26, B58, B62GRF9b proteinSSPDDQIGYKINSWITTIA2, B7, B8, B59, B58SSPDDQIGYA1, A26, B58, B62LPFSNVTWA1, A3, A26, B58, B62GRF9b proteinSSPDQIGYNINSQVAVLA2, B7, B58SSPDDQIGYA1, A26, B58, B62		AIVMVTIML	A2, B7		TELALINGT	A1 A2 A26 D59 D62	
EVVLKGVKLB7, B8VLRVNDKVYA1, A5, B62ESNRKFLPFA26, B62VLPFNDKVYA1, B62FAMQMAYRFB58, B62VLPFNDKVB7, B62FLNYTVPAA2, B8VYDPLQPELA24, B39FRKSNLKPFB8, B27WTGAAAQALA26, B58, B62FVFNLLPLA2, A26, B58, B62VLQPRIFLA26, B50, B62GAAAYYOGYA1, B58, B62YQDVNCTEVA1, A2, B39GAEHVNNSVA1, B62YQDVNCTEVA1, A2, B39, B62GQTGKIADYB27, B62YQDVNCTEVA1, A2, B39, B62GAIPINFTIA24, B58PUDTC9DLSPRWYFYA1, A3, A26GAIPINNTVB58, B62VVPAQEKNPA26, B62IDAVDCALA1, B39, B58USPRWYFYA1, A3, A26, B58, B62GGYQTSNFB58, B62KKNDLSPRWB58, B62KINDYNYKLA2, B39SPRWYFYYA1, A3, A26, B58, B62KINDYNYKLA2, B39SPRWYFYYA1, A3, A26, B58, B62KINDYCTMYA1, A3, B58, B62SPRDQIGYB8, B39KINDYCTMYA1, A3, B58, B62SPRDQIGYB8, B39KINDYCTMYA1, A3, B58, B62SPRWYFYYA1, A26, B58, B62KINDYNKLA2, B7B58, B62SPRWYFYYA1, A26, B58, B62KINDYCTMYA1, A3, B58, B62SPRWYFYYA1, A26, B58, B62KINDYCTMYA2, B7B58, B62SPRWYFYYA1, A26, B58, B62KINDYNKLA2, B7B58, B62SPRWYFYYA1, A26, B58, B62KINDYCKITMYA2, B7B58, B62SPRWYFYYA1, A26, B58, B62 <td></td> <td>DEDDSEPVL</td> <td>B39, B44</td> <td></td> <td>ISNQVAVLY</td> <td>A1, A3, A20, B58, B02</td>		DEDDSEPVL	B39, B44		ISNQVAVLY	A1, A3, A20, B58, B02	
ESNKKFLPF A26, B62 VLPNDGVY A1, B62 FAMQMAYRF B58, B62 VLPNDGVY A1, B62 FFVSQPFL B39, B44 VLPNQNAQAL B7, B62 FLVTYPA A2, B8 VTPPLQFEL A24, B39 FRKSNLKF B8, B27 VTOPLQFEL A24, B39 FKSSNLKF B8, B27 FTISVTTEI A2, A26, B58, B62 GAAAYYVGY A1, B58, B62 GAAAYYVGY A1, B58, B62 GQTGKLADY B27, B62 GQTGKLADY B58, B62 GQTGSLADY B58, B62 GQTGSLA		EPVLKGVKL	B7, B8		VLKGVKLHY	A1, A3, B62	
FAMQMAYRFB58, B62VRFNITNI,B27, B39FEYVSQFFLB39, B44VVNQNAQALB7, B62FLHVTYVPAA2, B8VYDPLQFELA24, B39FRKSNLKPFB8, B27WTGAGAAIA26, B58, B62FTISVTTEIA2, A26, B58WTGAGAAIA26, B58, B62GAAAYYVGYA1, B58, B62YQDVNCTEVA1, A26, B58, B62GAAAYYVGYA1, B58, B62YQDVNCTEVA1, A2, A39, B58, B62GQTGKLDPB27, B62YVPAQEKVTA2, A24, B8, B39, B62GQTGKLDPB27, B62YVPAQEKVTA2, A24, B8, B39, B62GQTGKLDPB27, B62SPRWYFYA1, A3, A26IAPTNFTIA24, B58PODTC9DLSFRWYFYA1, A3, A26, B58, B62IGAGICASYB58, B62KAYNVTQAFA24, B7, B8, B58, B62IGAVNDCALA1, B39, B58KKQTVTKLA2, B39KIADYNYKLA2, B39SRPWYFYYA1, A3, A26, B58, B62KIYSKHTPIA2, B39SPRWYFYLB7, B8KIYSKHTPIA2, B8SPRWYFYLB7, B8KIYSKDCTMYB7, B58SPROUGYA1, A26, B56LIALHRSYLA2, B62SPRWYFYLB7, B8NILLQYGSFB7, B8SEDEVEXFULA2, B7, B8, B39, B58, B62NGVEGFNCYA26, B62SEMHPALRLB39, B54, B62NILLQYGSFB8, B62SEMHPALRLB39, B54, B62NILLQYGSFB8, B62SEMHPALRLB39, B54, B62NISNQVAULA26, B58, B62SEMHPALRLB2, B53, B62NISNQVAULA26, B56SEMHPALRLB39, B54, B62		ESNKKFLPF	A26, B62		VLPFNDGVY	A1, B62	
FEY VSQPFLB39, B44VVNQNAQALB7, B62FLHVTYVPAA2, B8VTDPL/ELA24, B3FRKSNLKFFB8, B27WTGGAAAYYA1, A26, B58, B62FVISVTFEIA2, A26, B58WTGGGAALA26, B58, B62GAAAYYVGYA1, B58, B62YQPVRVVLA2, A28, B58, B62GAAAYYVGYA1, B58, B62YQPVRVVLA2, A24, B58, B62GAEHVNNSYA1, B62YQPKVVVLA2, A24, B58, B62GQTGKIADYB27, B62PODTC9DLSPRWYFYA1, A3, A26GAGICASYB58, B62KAYNV1QAFA24, B58, B62IDAVDCALA1, B39, B58B62KAYNV1QAFA24, B7, B8, B58, B62IDAVDCALA1, B39, B58, B62KGRYQTSNFB58, B62KIYSKHTPIA2, B3SSPDDQIGYA1, A3, A26, B58, B62KIYSKHTPIA2, B8SPRWYFYYLA1, A3, A26, B58, B62KIYSKHTPIA2, B8SPRWYFYYLB7, B8KIADYNYKLA2, B7, B8SSPDDQIGYA1, A26, B56, B62KVTLADAGFB58, B62GRF9b proteinKISEMHPALA2, B7, B8KISSNTWF7, B58KVYPIILRLA2, A3, B58B62LEPFFSNTWFF7, B58SEDKAFQLA2, B7, B3LLALHRSYLA26, B59SEMHPALRLB39, B54, B62QITTDNFFA24, A26, B58, B62SEMHPALRLB39, B54, B62NILLQYGSFB8, B62SEMHPALRLB39, B54, B62QUTTDYFKA24, A26, B58, B62SEMHPALRLB39, B54, B62QUTTDYFKA24, A26, B58, B62SEMHPALRLB39, B54, B62 <tr< td=""><td></td><td>FAMQMAYRF</td><td>B58, B62</td><td></td><td>VRFPNITNL</td><td>B27, B39</td></tr<>		FAMQMAYRF	B58, B62		VRFPNITNL	B27, B39	
FLHVTYVPAA2, B8YYDPLQPELA24, B39FRKSNLKPFB8, B27WTAGAAAYYA1, A26, B58, B62FTISVTTEIA2, A26, B8, B62YLQPRTFLLA2, B39, B58, B62GAAAYYVQYA1, B58, B62YQDVNCTEVA1, A2, B39, B58, B62GAEHVNNSYA1, B62YVPAQEKNFA2, A24, B8, B9, B62GQTGKIADYB27, B62YVPAQEKNFA2, 662GQTGKIADYB58, B62YVPAQEKNFA2, 662IAIPTNFTIA24, B58PODTC9DLSPRWYFYA1, A3, A26IGAGICASYB58, B62KAYNVTQAFA24, B7, B8, B88, B62IGIVNNTVYB58, B62KAYNVTQAFA24, B7, B8, B58, B62ITDAVDCALA1, B39, B58GFAPSASAFA24, B62KIADYNYKLA2, B3GFAPSASAFA24, B62KINYSKHTPIA2, B8, B62OFAPSASAFA24, B62, B62KINYSKHTPIA2, B8, B62OPDTD2GFAVSASFA24, B62, B62KINYSKHTPIA2, B8, B62OPDTD2GFKVYPILB7, B8KIADYNYKLA2, B8, B62OPDTD2GFKVYPILB7, B8, B62KINTSNCTIMYA1, A3, B58, B62OPDTD2GFKVYPILB7, B8, B63LSETKCTLKA1, A3RLNDPQIQLA2, B7, B8, B39, B58, B62MTSCCSCLKA1, A3RLNDPQIQLA2, B7, B8, B39, B44SLEDKAFQLA2, B7, B58, B62SEMIPALRI, B39, B44NILLQYGSFB8, B62SEMIPALRI, B39, B44NLLLQYGSFB8, B62Significant pMHC-TCR interactions (Fig. 1), This hit pep-QITTDNTFA24, A26, B58, B62Tom influe		FEYVSQPFL	B39, B44		VVNQNAQAL	B7, B62	
FRKSNLKPFB8, B27WTAGAAAYYA1, A26, B58, B62FTISVTTEIA2, A26, B58WTFGAGAALA26, B62FVFLVLLPLA2, A26, B8, B62YQPKTVVLA2, B39, B58, B62GAAAYY VGYA1, B58, B62YQPYRVVVLA2, A24, B8, B39, B62GAEHVNNSYA1, B62YQPYRVVVLA2, A24, B5, B39, B62GAEIVNNSYB58, B62YVPAQEKNFA26, B62GAGICASYB58, B62VVPAQEKNFA26, B58, B62IGIVNNTVYB58, B62KMKDLSFRWB58, B62IGIVNNTVYB58, B62KMKDLSRWB58, B62KIADYNYKLA2, B3CKKQQTVLB8, B59, B62KIADYNYKLA2, B8CPAPSASAFA24, B62KIADYNYKLA2, B8CPAPSASAFA24, B62KUTLADAGFB58, B62ORF9b proteinGPKVYPILB7, B8KUTLADAGFB58, B62ORF9b proteinKISEMHPLA2, B7, B8, B39, B58, B62LALHRSYLA1, A3, B58, B62SSPDDQICYA1, A2, B56LSFTKCTLKA1, A3KISEMHPLA2, B7, B8, B39, B58, B62LSFTKCTLKA1, A3KVYPILRLA2, A3, B58LSFTKCTLKA1, A3RLVDPQIQLA2, B62NTSNQVAVLA26, B58, B62SEMHPALRLB39, B44SLEDKAFQLA2, B7, B8, B58, B62SEMHPALRLB39, B44NLLLQYGSFB8, B62SEMHPALRLB39, B44SLEDKAFQLA2, B7, B8, B62SEMHPALRLB39, B44SLEDKAFQLA2, B7, B8, B62SEMHPALRLB39, B44SLEDKAFQLA2, B7, B8, B62<		FLHVTYVPA	A2, B8		VYDPLQPEL	A24, B39	
FITSVTTEIA2, A26, B58WTFGAGAALA26, B62FVFLVLLPLA2, A26, B8, B62YLQPRTFLLA2, B39, B58, B62GAAAYYVQYA1, B58, B62YQDVNCTEVA1, A2, B39GAEHVNNSYA1, B62YQPYRVVULA2, A24, B8, B39, B62GQTGKIADYB27, B62YUPAQEKNFA26, B62LAIPTNFTIA24, B58PDDTC9DLSPRWYFYA1, A3, A26IGAGICASYB58, B62FPRQGQVHB7, B8IGAGICASYB58, B62KAYNVTQAFA24, B7, B8, B58, B62IGIVNNTVYB58, B62KMKDLSPRWB58, B62IGIVNNTVKA2, B39QRKKQQTVTLB8, B39KIADYNYKLA2, B3CPAPSASAFA24, B62KIYSKHTPIA2, B8SPDDQIGYA1, A3, A26, B58, B62KIYSKHTPIA2, B8SPRWYFYYLB7, B8LLALIRSYLA2, B8SPDDQIGYA1, A26, B62LALLIRSYLA2, B8SPDDQIGYA1, A26, B62LALILRYXLA2, B7, B8, B58, B62CORP9b proteinGFKVYPIILRTSCCSCLKA1, A3MARKTLNSLB7, B8MTSCCSCLKA1, A3MARKTLNSLB7, B8MTSQVAVLA26, B59SEMHPALRLB39, B44SLEDKAFQLA2, B7, B58, B62SEMHPALRLB39, B44SLEDKAFQL <td></td> <td>FRKSNLKPF</td> <td>B8. B27</td> <td></td> <td>WTAGAAAYY</td> <td>A1, A26, B58, B62</td>		FRKSNLKPF	B8. B27		WTAGAAAYY	A1, A26, B58, B62	
FIGUREA2, A26, B8, B62YLQPRTFLLA2, B39, B58, B62GAAAYYVGYA1, B58, B62YQDVNCTEVA1, A2, B39GAEHNNSYA1, B62YQPYRVVULA2, A24, B8, B39, B62GQTGKIADYB27, B62YVPAQEKNFA26, B62IAIPTNFTIA24, B58PODTC9DLSPRWYFYA1, A3, A26IGAGICASYB58, B62FPRGQGVPIB7, B8IGAYOTXVB58, B62KAYNYTQAFA24, B7, B8, B58, B62IGIVNNTVYB58, B62KKINDLSPRWB58, B62ITDAVDCALA1, B39, B58GPAFSASAFA24, B7, B8, B58, B62KIADYNYKLA2, B39QRKKQTVTIB8, B39KIADYNYKLA2, B8SSPDDQIGYA1, A26, B62KINSKHTPIA2, B8SSPDDQIGYA1, A26, B62KVTLADAGFB58, B62PODTD2GPKVYPILB7, B8LEFFENNTWB7, B58B62SSPDDQIGYA1, A26, B62LPFFSNTWB7, B58B62SEMHPALRLA2, B7, B8, B39, B58, B62NGVEGFNCYA26, B62SEMHPALRLB39, B44SLEDKAFQLA2, B7, B8, B62SEMHPALRLB39, B44NLLQYGSFB8, B62SEMHPALRLB39, B44NLLQYGSFB8, B62SEMHPALRLB39, B44SLEDKAFQLA2, B7, B8, B62SEMHPA		FTISVTTFI	A2 A26 B58		WTFGAGAAL	A26, B62	
InterfactAL, AG, BB, BG2YQDVNCTEVA1, A2, B39GAAAYYVGYA1, B58, B62YQPYRVVUA2, A24, B8, B39, B62GQTGKIADYB27, B62YVPAQEKNFA26, B62IAIPTNFTIA24, B58PODTC9DLSPRWYFYA1, A3, A26IGAGICASYB58, B62KAYNYCAFA24, B7, B8, B58, B62IGIVNNTVYB58, B62KAYNYCAFA24, B7, B8, B58, B62IGIVNNTVYB58, B62KAYNYFYYA1, A3, A26, B58, B62KIADYNYKLA2, B39QRQKKQQTVB8, B39KIADYNYKLA2, B3QRQKKQQTVB8, B39KIYSKHTPIA2, B8SSPDDOIGYA1, A26, B62KVTLADAGFB58, B62PODTD2GPKVYPILB7, B8KIYSKHTPIA2, B8SSPDOIGYA1, A26, B62LAHRSYLA2, B8SSPDOIGYA1, A26, B62LIALHRSYLA2, B8SSPDOIGYA1, A26, B62LIALHRSYLA2, B8SSPDOIGYA1, A26, B62SISTEKCTLKA1, A3MARKTLNSLB7, B8MTSCCSCLKA1, A3MARKTLNSLB7, B8NTSNQVAVLA26, B62SEMHPALRLB39, B44NLLQYGSFB8, B62SEMHPALRLB39, B44QLTPTWRVYA1, B62Ioga), which is a linear peptide cpitope of matrix protein 1RVVVLSFELA2, B7, B8, B62Significant pMHC-TCR interactions (Fig. 1). This hit peptide is portein influenza A virus as recorded in IEDB and shows 40SILAYTMSLA2, A5, B58, B62Significant pMHC-TCR interactions (Fig. 1). This hit peptide is portide or interactions (Fig. 1). This hit pep			A2 A26 D8 D62		YLQPRTFLL	A2, B39, B58, B62	
GAAATIYNGI AI, B62 GAEHVNNSY AI, B62 GQTGKIADY B27, B62 IAIPTNFTI A24, B58 IGAGICASY B58, B62 IGIVNNTVY B58, B62 ITDAVDCAL AI, B39, B58 KGIYQTSNF B58, B62 KIADYNYKL A2, B39 KIADYNYKL A2, B39 KIADYNYKL A2, B39 KIXDYNYKL A2, B38 KTSVDCTMY A1, A3, B58, B62 LIFFSNVTW B7, B58 LLALHRSYL A2, B8 LLALHRSYL A2, B8 LISETKCTLK A1, A3 MTSCCSCLK A1, A3 MTSCCSCCLK A1, A3 M			A2, A20, B6, B02		YQDVNCTEV	A1, A2, B39	
GAEHVINSYA1, B62YUPAQEKNFA26, B52GQTGKLADYB27, B62PODTC9DLSPRWYFYA1, A3, A26IAIPTNFTIA24, B58PODTC9PRGQGVPIB7, B8IGAGICASYB58, B62KAYNVTQAFA24, B7, B8, B58, B62IGIVNNTVYB58, B62KMKDLSPRWB58, B62ITDAVDCALA1, B39, B58GACQTVTLB8, B39KIADYNYKLA2, B39GKKQQTVTLB8, B39KIADYNYKLA2, B8GPAPSASAFA24, B7, B8, B39, B58, B62KIYSKHTPIA2, B8SPRWYFYYLB7, B8KTSVDCTMYA1, A3, B58, B62PODTD2GFKVYPIILRVTLADAGFB58, B62PODTD2GFKVYPIILRVTLADAGFB58, B62PODTD2GFKVYPIILRVTSCCSCLKA1, A3MARKTLNSLA2, B62SEMHPALA2, B7, B8MTSCCSCLKA1, A3RLVDPQIQLA2, B62NTSNQVAVLA26, B62SEMHPALRLB39, B44NLLLQYGSFB8, B62SLEDKAFQLA2, B39QITTDNTFA24, A26, B58, B62significant pMHC-TCR interactions (Fig. 1). This hit pep-QLTPTWRVYA1, B62SIGNOSUS to template peptide GILGFVFTL (PDB)QUITTNNTFA2, A26, B62SIGNIGANT A26, B62SILAYTMSLA2, A26, B62SIGNIGANT A26, B62SILAYTMSLA2, A26, B62SIGNIGANT A26, B62SILAYTMSLA2, A26, B62SIGNIGANT A26, B62SILAYTMSLA2, B7, B6SICOCOP (A26, B62SILAYTMSLA2, A26, B62SIGNIGANT A26, B62SILAYTMSL </td <td></td> <td>GAAATIVGT</td> <td>A1, B38, B02</td> <td></td> <td>YOPYRVVVL</td> <td>A2, A24, B8, B39, B62</td>		GAAATIVGT	A1, B38, B02		YOPYRVVVL	A2, A24, B8, B39, B62	
GQTGKIADYB27, B62DDTC9DLSPRWYFYAI, A3, A26IAIPTNFTIA24, B58PODTC9Nucleoprotein (N)DLSPRWYFYAI, A3, A26IGAGICASYB58, B62KAYNVTQAFA24, B7, B8, B58, B62IGIVNNTVYB58, B62LSPRWYFYYAI, A3, A26, B58, B62ITDAVDCALA1, B39, B58SPRWYFYYAI, A3, A26, B58, B62KIYSKHTPIA2, B39QRQKKQQTVB8, B39KIYSKHTPIA2, B8SSPDDQIGYAI, A26, B62KVTLADAGFB58, B62PODTD2GPKVYPILB7, B8SSPDDQIGYAI, A26, B62LLALHRSYLA2, B8SSPDDQIGYAI, A26, B62LLALHRSYLA2, B8KVYPIILRLA2, A3, B58LSETKCTLKA1, A3MAKKTLNSLB7, B8MTSCCSCLKA1, A3MAKKTLNSLB7, B8NULLQYGSFB8, B62SEMHPALRLB39, B44NLLLQYGSFB8, B62Significant pMHC-TCR interactions (Fig. 1). This hit pep-UITTDNTFA24, A26, B58, B62Significant pMHC-TCR interactions (Fig. 1). This hit pep-UITTDNTFA24, A26, B58, B62Significant pMHC-TCR interactions (Fig. 1). This hit pep-UITTDNTFA24, A26, B58, B62Significant pMHC-TCR interactions (Fig. 1). This hit pep-UITTDNTFA24, A26, B58, B62Significant pMHC-TCR interactions (Fig. 1). This hit pep-UITTDNTFA24, A26, B58, B62Significant pMHC-TCR interactions (Fig. 1). This hit pep-UITTONTFA24, A26, B58, B62Significant pMHC-TCR interactions (Fig. 1). This hit pep-UITTONTFA24, A		GAEHVNNSY	A1, B62		YVPAOEKNF	A26. B62	
IAIPTNFTIA24, B58District of the periode and		GQTGKIADY	B27, B62	ΡΟΟΤΟ9	DI SPRWYFY	A1 A3 A26	
IGAGICASYB58, B62KAYNVTQAFA24, B7, B8, B58, B62IGIVNNTVYB58, B62KMKDLSPRWB58, B62ITDAVDCALA1, B39, B58LSPRWYFYYA1, A3, A26, B58, B62KGIYQTSNFB58, B62QFAPSASAFA24, B62KIADYNYKLA2, B39QRQKKQQTVB8, B39KIYSKHTPIA2, B8SPRWYFYYLB7, B8KTSVDCTMYA1, A3, B58, B62QFKVYFILB7, B8KVTLADAGFB58, B62QRF9b proteinGPKVYPILRVVTLADAGFB58, B62QRF9b proteinKISEMHPALLALHRSYLA2, B8SPDQIGYA1, A26, B62LSETKCTLKA1, A3MARKTLNSLB7, B8LSETKCTLKA1, A3MARKTLNSLB7, B8MTSCCSCLKA1, A3MARKTLNSLB7, B8NTSNQVAVLA26, B62SEMHPALRLB39, B44NLLLQYGSFB8, B62Significant pMHC-TCR interactions (Fig. 1). This hit pep-QLTPTWRVYA1, B62Ioga), which is a linear peptide GILGFVFTL (PDB:QLTPTWRVYA1, B62Significant pMHC-TCR interactions (Fig. 1). This hit pep-QLTPTWRVYA1, B62Significant pMHC-TCR interactions (Fig. 1). This hit protein 1RVVVLSFELA2, B7, B58, B62Significant pMHC-TCR interactions (Fig. 1). This hit pep-SILASTASALA2, B7, B62Significant pMHC-TCR interactions (Fig. 1). This hit pep-SILASTASALA2, B7, B62Significant pMHC-TCR interactions (Fig. 1). This hit pep-SILASTASALA2, B7, B62Significant pMHC-TCR interactions (Fig. 1). This hit pep-SILAST		IAIPTNFTI	A24, B58	Nucleoprotein (N)	FPRGQGVPI	B7, B8	
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ITDAVDCALA1, B39, B58LSPRWYFYYA1, A3, A26, B58, B62KGIYQTSNFB58, B62QKKQQTVTLB8, B39KIADYNYKLA2, B39QRQKKQUTV B8, B27KIYSKHTPIA2, B8SPRWYFYLB7, B8KTSVDCTMYA1, A3, B58, B62P0DTD2GPKVYPILKVTLADAGFB58, B62P0DTD2GPKVYPILB7, B58CRF9b proteinKISEMHPALA2, B7, B8, B39, B58, B62LLALHRSYLA2, B8KVYPIILRLA2, A3, B58LSETKCTLKA1, A3MARKTLNSLB7, B8MTSCCSCLKA1, A3RLVDPQIQLA2, B62NGVEGFNCYA26, B62SEMHPALRLB39, B44NLLLQYGSFB8, B62SLEDKAFQLA2, B39QIITTDNTFA24, A26, B58, B62significant pMHC-TCR interactions (Fig. 1). This hit pep-QLTPTWRVYA1, B62riga), which is a linear peptidic epitope of matrix protein 1RVVVLSFELA2, B7, B58, B62riga in peptidic antigen family of template loga acrossSLSSTASALA2, B7, B6225 organisms.The peptide set from antigen P0DTC9 has also shown the same hit peptide for all the four target populations. The same hit peptide for all the four target populations. The same hit peptide for all the four target populations. The same hit peptide for all the four target populations. The same hit peptide for all the four target populations. The same hit peptide for all the four target populations. The same hit peptide for all the four target populations. The same hit peptide for all the four target populations. The same hit peptide for all the four target populations. The same hit peptide for all the four target populations. The same hit peptide for al		IGIVNNTVY	B58, B62		KMKDLSPRW	B58, B62	
KGIYQTSNFB58, B62QFAPSASAFA24, B62KIADYNYKLA2, B39QRQKKQQTVLB8, B39KIYSKHTPIA2, B8QRQKKQQTVB8, B27KIYSKHTPIA2, B8SPRWYFYLB7, B8KTSVDCTMYA1, A3, B58, B62PODTD2GPKVYPILKVTLADAGFB58, B62ORF9b proteinKISEMHPALA2, B7, B58LLALHRSYLA2, B8B62LPFFSNVTWB7, B58KIRLGSPLSLB27, B39LSETKCTLKA1, A3MARKTLNSLB7, B8MTSCCSCLKA1, A3RLVDPQIQLA2, B62NGVEGFNCYA26, B62SEMHPALRLB39, B44NLLLQYGSFB8, B62SLEDKAFQLA2, B39QITTDNTFA24, A26, B58, B62significant pMHC-TCR interactions (Fig. 1). This hit pep-QLTPTWRVYA1, B62significant pMHC-TCR interactions (Fig. 1). This hit pep-QLTPTWRVYA1, B62Significant pMHC-TCR interactions (Fig. 1). This hit pep-UITTDNTFA24, A26, B58, B62significant pMHC-TCR interactions (Fig. 1). This hit pep-UITTYMSLA2, A26, B62Significant pMHC-TCR interactions (Fig. 1). This hit pep-UITTYMSLA2, A26, B62Significant pMHC-TCR interactions (Fig. 1). This hit pep-UITYWXYA1, B62Significant pMHC-TCR interactions (Fig. 1). This hit pep-UITYTMSLA2, A26, B62Significant pMHC-TCR interactions (Fig. 1). This hit pep-SILAYTMSLA2, A26, B62Significant pMHC-TCR interactions (Fig. 1). This hit pep-SILAYTMSLA2, A26, B62Significant pMHC-TCR interactions (Fig.		ITDAVDCAL	A1, B39, B58		LSPRWYFYY	A1, A3, A26, B58, B62	
KIADYNYKLA2, B39QRNQUTVILB8, B39KIYSKHTPIA2, B8SPRWYFYYLB7, B8KTSVDCTMYA1, A3, B58, B62PODTD2GPKVYPILB7, B8KVTLADAGFB58, B62PODTD2GPKVYPILB7, B8LLALHRSYLA2, B8RESPEDQIGYA1, A26, B62LSETKCTLKA1, A3KVYPIILRLA2, A3, B58LSETKCTLKA1, A3RLVDPQIQLA2, B62NGVEGFNCYA26, B62SEMHPALLB39, B44NLLLQYGSFB8, B62SLEDKAFQLA2, B39QIITTDNTFA24, A26, B58, B62significant pMHC-TCR interactions (Fig. 1). This hit pep-QLTPTWRVYA1, B62from influenza A virus as recorded in IEDB and shows 40SILAYTMSLA2, A26, B62SE from antigen P0DTC9 has also shownSPRRARSVAB7, B8The peptide set from antigen P0DTC9 has also shownSPRRARSVAB7, B8The peptide for all the four target populations. The		KGIYQTSNF	B58, B62		QFAPSASAF	A24, B62	
KIYSKHTPIA2, B8SPRWYFYYLB7, B8KTSVDCTMYA1, A3, B58, B62PDDTD2GFKVYPILB7, B8KVTLADAGFB58, B62PDDTD2GFKVYPILB7, B8LLALHRSYLA2, B8B62RVYPIILRLA2, B7, B8, B39, B58, B62LPFFSNVTWB7, B58LLGSPLSLB27, B39LSETKCTLKA1, A3MARKTLNSLB7, B8MTSCCSCLKA1, A3MLVDPQIQLA2, B62NGVEGFNCYA26, B62SEMHPALRLB39, B44NLLLQYGSFB8, B62SLEDKAFQLA2, B39QIITTDNTFA24, A26, B58, B62significant pMHC-TCR interactions (Fig. 1). This hit pep-QLTPTWRVYA1, B62significant pMHC-TCR interactions (Fig. 1). This hit pep-QLTPTWRVYA1, B62significant pMHC-TCR interactions (Fig. 1). This hit pep-QLTPTWRVYA1, B62significant pMHC-TCR interactions (Fig. 1). This hit pep-IGBA, WICH SALLA2, A26, B58, B62SIGNIFICAN A virus as recorded in IEDB and shows 40SILAYTMSLA2, A26, B62peptides in peptide antigen family of template loga acrossSLSSTASALA2, B7, B62The peptide set from antigen PODTC9 has also shown the same hit peptide for all the four target populations. The senter hit peptide for all the four target populations. The senter hit peptide for all the four acregit populations. The senter hit peptide for all the four target populations. The senter hit peptide for all the four target populations. The		KIADYNYKL	A2, B39		OPOKKOOTV	B8, B39 B8, B27	
KTSVDCTMYA1, A3, B58, B62SSPDDQGYA1, A26, B62KVTLADAGFB58, B62PDTD2GPKVYPIILB7, B8LLALHRSYLA2, B8CRF9b proteinKISEMHPALA2, B7, B8, B39, B58, B62LPFFSNVTWB7, B58KVYPIILRLA2, A3, B58LSETKCTLKA1, A3KVVPIILRLA2, A3, B58MTSCCSCLKA1, A3RLVDPQIQLA2, B62NGVEGFNCYA26, B62SEMHPALRLB39, B44NLLLQYGSFB8, B62SLEDKAFQLA2, B39QITTTDNTFA24, A26, B58, B62significant pMHC-TCR interactions (Fig. 1). This hit pep- tide is homologous to template peptide GILGFVFTL (PDB: loga), which is a linear peptidic epitope of matrix protein 1 from influenza A virus as recorded in IEDB and shows 40 peptides in peptide antigen family of template loga across 25 organisms. The peptide for all the four target populations. The entope of the peptide for all the four target populations. The entope of the peptide for all the four target populations. The entope of the paper found to have a pint Z		KIYSKHTPI	A2. B8		SPRWYFYYL	B7, B8	
KTSVDCTMTAT, AS, DS, B02KVTLADAGFB58, B62KVTLADAGFB58, B62LLALHRSYLA2, B8LPFFSNVTWB7, B58LSETKCTLKA1, A3MTSCCSCLKA1, A3MTSCCSCLKA1, A3MTSCCSCLKA1, A3MTSCCSCLKA1, A3MCVEGFNCYA26, B62NGVEGFNCYA26, B62NTSNQVAVLA26, B39QIITTDNTFA24, A26, B58, B62QLTPTWRVYA1, B62QLTPTWRVYA1, B62SILAYTMSLA2, A26, B62SILAYTMSLA2, B7, B58, B62SILAYTMSLA2, B7, B58, B62SILAYTMSLA2, B7, B8STECSNLLLA1, B39		KTSVDCTMV	A1 A2 D58 D62		SSPDDQIGY	A1, A26, B62	
KV1LADAGFB58, B62ORF9b proteinKISEMHPALA2, B7, B8, B39, B58, B62LLALHRSYLA2, B7, B58KVYPIILRLA2, A3, B58LPFFSNVTWB7, B58LRLGSPLSLB27, B39LSETKCTLKA1, A3MARKTLNSLB7, B8MTSCCSCLKA1, A3RLVDPQIQLA2, B62NGVEGFNCYA26, B62SEMHPALRLB39, B44NLLLQYGSFB8, B62SLEDKAFQLA2, B39QITTTDNTFA24, A26, B58, B62significant pMHC-TCR interactions (Fig. 1). This hit pep-QLTPTWRVYA1, B62loga), which is a linear peptide GILGFVFTL (PDB:loga), which is a linear peptide of matrix protein 1from influenza A virus as recorded in IEDB and shows 40SILAYTMSLA2, A26, B62peptides in peptide antigen family of template loga acrossSLSSTASALA2, B7, B825 organisms.SPRRARSVAB7, B8The peptide set from antigen P0DTC9 has also shownSTECSNLLLA1, B39A1, B39			A1, A3, B36, B02	P0DTD2	GPKVYPIIL	B7, B8	
LPFFSNVTWB7, B58KVYPIILRLA2, A3, B58LSETKCTLKA1, A3LRLGSPLSLB27, B39MTSCCSCLKA1, A3MARKTLNSLB7, B8MTSCCSCLKA1, A3RLVDPQIQLA2, B62NGVEGFNCYA26, B62SEMHPALRLB39, B44NLLLQYGSFB8, B62SLEDKAFQLA2, B39NTSNQVAVLA26, B39Significant pMHC-TCR interactions (Fig. 1). This hit pep- tide is homologous to template peptide GILGFVFTL (PDB: loga), which is a linear peptidic epitope of matrix protein 1 from influenza A virus as recorded in IEDB and shows 40SILAYTMSLA2, A26, B62peptide antigen family of template loga across 25 organisms. The peptide for all the four target populations. The tes ame hit peptide for all the four target populations. The the same hit peptide for all the four target populations. The tes met hit peptide for all the four target populations. The tes met hit peptide for all the four target populations. The tes met hit peptide for all the four target populations. The tes met hit peptide for all the four target populations. The tes met hit peptide for all the four target populations. The tes met hit peptide for all the four target populations. The tes met hit peptide for all the four target populations. The tes met hit peptide for all the four target populations. The tes met hit peptide for all the four target populations. The tes met hit peptide for all the four target populations. The tes met hit peptide for all the four target populations. The tes met hit peptide for all the four target populations. The tes met hit peptide for all the four target populations. The tes met hit peptide for all the four target populations. The tes met hit peptide for all the four target populations. The <td></td> <td>K V I LADAGF LLALHRSYL</td> <td>A2, B8</td> <td>ORF9b protein</td> <td>KISEMHPAL</td> <td>A2, B7, B8, B39, B58, B62</td>		K V I LADAGF LLALHRSYL	A2, B8	ORF9b protein	KISEMHPAL	A2, B7, B8, B39, B58, B62	
LRLGSPLSL B27, B39 LSETKCTLK A1, A3 MTSCCSCLK A1, A3 MTSCCSCLK A1, A3 MTSCCSCLK A1, A3 MTSCCSCLK A1, A3 MTSCCSCLK A1, A3 MARKTLNSL B7, B8 RLVDPQIQL A2, B62 SEMHPALRL B39, B44 SLEDKAFQL A2, B39 NTSNQVAVL A26, B39 QIITTDNTF A24, A26, B58, B62 QLTPTWRVY A1, B62 QLTPTWRVY A1, B62 SIIAYTMSL A2, A26, B62 SLSSTASAL A2, B7, B68 SLSSTASAL A2, B7, B62 SPRRARSVA B7, B8 STECSNLLL A1, B39 LRLGSPLSL B27, B39 MARKTLNSL B7, B3 SIGNIFICAT PMHC-TCR interactions (Fig. 1). This hit pep- tide is homologous to template peptide GILGFVFTL (PDB: loga), which is a linear peptidic epitope of matrix protein 1 from influenza A virus as recorded in IEDB and shows 40 peptides in peptide antigen family of template loga across 25 organisms. The peptide set from antigen P0DTC9 has also shown the same hit peptide for all the four target populations. The environe SPRWYEVYU has been found to have a joint 7		I DEESNIVTW	B7 B58		KVYPIILRL	A2, A3, B58	
LSEIRCTENAI, A3MARKTLNSLB7, B8MTSCCSCLKA1, A3RLVDPQIQLA2, B62NGVEGFNCYA26, B62SEMHPALRLB39, B44NLLLQYGSFB8, B62SLEDKAFQLA2, B39NTSNQVAVLA26, B39Significant pMHC-TCR interactions (Fig. 1). This hit pep-QLTPTWRVYA1, B62significant pMHC-TCR interactions (Fig. 1). This hit pep-QLTPTWRVYA1, B62significant pMHC-TCR interactions (Fig. 1). This hit pep-RVVVLSFELA2, B7, B58, B62from influenza A virus as recorded in IEDB and shows 40SILAYTMSLA2, A26, B62peptides in peptide antigen family of template loga acrossSLSSTASALA2, B7, B62Z5 organisms.SPRRARSVAB7, B8The peptide for all the four target populations. TheSTECSNLLLA1, B39an ipptide for all the four target populations. The			A1 A2		LRLGSPLSL	B27, B39	
MTSCCSCLKA1, A3RLVDPQIQLA2, B62NGVEGFNCYA26, B62SEMHPALRLB39, B44NLLLQYGSFB8, B62SLEDKAFQLA2, B39NTSNQVAVLA26, B39Significant pMHC-TCR interactions (Fig. 1). This hit pep- tide is homologous to template peptide GILGFVFTL (PDB: loga), which is a linear peptidic epitope of matrix protein 1 from influenza A virus as recorded in IEDB and shows 40SILAYTMSLA2, A26, B62peptides in peptide antigen family of template loga across 25 organisms. The peptide set from antigen P0DTC9 has also shown the same hit peptide for all the four target populations. The enitope SPRWYEYYL has been found to have a joint 7		LSEIKCILK	A1, A3		MARKTLNSL	B7, B8	
NGVEGFNCYA26, B62SEMHPALRLB39, B44NLLLQYGSFB8, B62SLEDKAFQLA2, B39NTSNQVAVLA26, B39significant pMHC-TCR interactions (Fig. 1). This hit pep- tide is homologous to template peptide GILGFVFTL (PDB: loga), which is a linear peptidic epitope of matrix protein 1 from influenza A virus as recorded in IEDB and shows 40SILAYTMSLA2, A26, B62peptides in peptide antigen family of template loga acrossSILAYTMSLA2, B7, B62Steres and the four target populations. The apitope SPRWYEYYL has been found to have a joint 7		MTSCCSCLK	A1, A3		RLVDPQIQL	A2, B62	
NLLLQYGSFB8, B62SLEDKAFQLA2, B39NTSNQVAVLA26, B39Significant pMHC-TCR interactions (Fig. 1). This hit pep- tide is homologous to template peptide GILGFVFTL (PDB: loga), which is a linear peptidic epitope of matrix protein 1 from influenza A virus as recorded in IEDB and shows 40 peptides in peptide antigen family of template loga across 25 organisms. The peptide set from antigen P0DTC9 has also shown the same hit peptide for all the four target populations. The aritope SPRWYEYVL has been found to have a joint 7		NGVEGFNCY	A26, B62		SEMHPALRL	B39, B44	
NTSNQVAVLA26, B39QIITTDNTFA24, A26, B58, B62significant pMHC-TCR interactions (Fig. 1). This hit pep- tide is homologous to template peptide GILGFVFTL (PDB: 10ga), which is a linear peptidic epitope of matrix protein 1 from influenza A virus as recorded in IEDB and shows 40 peptides in peptide antigen family of template 10ga across 25 organisms. The peptide set from antigen P0DTC9 has also shown the same hit peptide for all the four target populations. The epitope SPRWYEYYL has been found to have a joint 7		NLLLQYGSF	B8, B62		SLEDKAFQL	A2, B39	
QIITTDNTFA24, A26, B58, B62significant pMHC-TCR interactions (Fig. 1). This hit pep- tide is homologous to template peptide GILGFVFTL (PDB: loga), which is a linear peptidic epitope of matrix protein 1 from influenza A virus as recorded in IEDB and shows 40 peptides in peptide antigen family of template loga across 25 organisms. The peptide set from antigen P0DTC9 has also shown the same hit peptide for all the four target populations. The apitope SPRWYEXYL has been found to have a joint 7		NTSNQVAVL	A26, B39				
QLTPTWRVYA1, B62tide is homologous to template peptide GILGFVFTL (PDB: loga), which is a linear peptidic epitope of matrix protein 1 from influenza A virus as recorded in IEDB and shows 40SIIAYTMSLA2, A26, B62peptides in peptide antigen family of template loga acrossSLSSTASALA2, B7, B6225 organisms.SPRRARSVAB7, B8The peptide set from antigen P0DTC9 has also shown the same hit peptide for all the four target populations. The epitope SPRWYEYYL has been found to have a joint 7		QIITTDNTF	A24, A26, B58, B62	significant pMHC-	TCR interactions	(Fig. 1). This hit pep-	
RVVVLSFELA2, B7, B58, B62from influenza A virus as recorded in IEDB and shows 40SIIAYTMSLA2, A26, B62peptides in peptide antigen family of template loga acrossSLSSTASALA2, B7, B6225 organisms.SPRRARSVAB7, B8The peptide for all the four target populations. The enitope SPRWYEYYL has been found to have a joint 7		QLTPTWRVY	A1, B62	tide is homologous	le GILGFVFTL (PDB:		
SIIAYTMSLA2, A26, B62peptides in peptide antigen family of template loga acrossSLSSTASALA2, B7, B6225 organisms.SPRRARSVAB7, B8The peptide set from antigen P0DTC9 has also shown the same hit peptide for all the four target populations. The enitone SPRWYEVYL has been found to have a joint 7		RVVVLSFEL	A2, B7, B58, B62	from influenza A x	n IEDB and shows 40		
SLSSTASALA2, B7, B6225 organisms.SPRRARSVAB7, B8The peptide set from antigen P0DTC9 has also shown the same hit peptide for all the four target populations. The epitope SPRWYEVYL has been found to have a joint 7		SIIAYTMSL	A2, A26, B62	peptides in peptide antigen family of template 10			
SPRRARSVAB7, B8The peptide set from antigen P0DTC9 has also shown the same hit peptide for all the four target populations. The epitope SPRWXEXVL has been found to have a joint 7		SLSSTASAL	A2, B7, B62	25 organisms.			
the same hit peptide for all the four target populations. The spitope SPRWYEVYL has been found to have a joint 7		SPRRARSVA	B7. B8	The peptide set	DTC9 has also shown		
		STECSNLLL	A1, B39	the same hit peptid	le for all the four to YYL has been for	arget populations. The und to have a joint \mathbf{Z}	

STQDLFLPF A1, A24, A26, B62

Table 4 Population coverage analysis of optimized top scoring CTL epitopes for different target populations different target populations	Protein	Target population	Epitope	Percentage coverage	Total HLA hits
	P0DTC2	Europe	RVVVLSFEL	80.55%	31
	Spike glycoprotein (S)	-	SLSSTASAL	77.12%	31
			YQPYRVVVL	75.44%	24
			AIVMVTIML	72.91%	18
			FVFLVLLPL	68.03%	20
			TSNQVAVLY	63.35%	26
			YLQPRTFLL	63.31%	26
			SIIAYTMSL	60.53%	33
		North America	RVVVLSFEL	80.83%	31
			SLSSTASAL	80.83%	31
			SIIAYTMSL	78.10%	33
			YOPYRVVVL	74.61%	24
			AIVMVTIML	69.55%	18
			TSNOVAVLY	67.13%	26
			VVNONAOAL	64.63%	23
			YLOPRTFLL	64.56%	26
		South Asia	TSNOVAVLY	80.64%	26
			KTSVDCTMY	76.42%	23
			VIKGVKLHY	71.64%	17
			LSETKCTLK	66 63%	10
			MTSCCSCLK	66 63%	10
			TLLALHRSY	66 62%	13
			SILAYTMSI	65.03%	33
			RVVVI SFEL	61 94%	31
		North East Asia	TSNOVAVLY	82.88%	26
			KTSVDCTMY	81.61%	23
			RVVVI SFFI	79.33%	31
			VI KGVKI HY	78.68%	17
			TLLALHRSY	76.03%	13
			SUSSTASAI	73.78%	31
			VVNONAOAI	68.07%	23
			SILAVTMSI	66 22%	33
	P0DTC9 Nucleoprotein (N)	Europe	KAYNVTOAF	77 43%	27
			I SPRWYFYY	63 35%	26
	1		SPRWYFYYI	60.10%	14
			FPRGOGVPI	59.75%	17
		North America	KAVNVTOAE	76 80%	27
		North America	ISPRWVEVV	67 13%	27
			DI SPRWVEV	54%	13
			SDRWVEVVI	51 20%	13
		South Asia	I SDDWVEVV	S1.20%	14 26
		South Asia	DI SDDWVEV	80.04 %	12
			KAVNWTOAE	72.00%	13
			KATINV I QAF SDDWVEVVI	03.14% 32.00%	27 14
		North Fost Asia	JENWIFIIL	52.77%	14
		North East Asia	LOFKW IFYY MAVNIJTOAE	0∠.00% 76.07%	20
			NATINY IQAF	10.21%	12
			DLOPKW IFI	04%	13
			SPKWYFYYL	25.03%	14

Table 4 (continued)	Protein	Target population	Epitope	Percentage coverage	Total HLA hits
	P0DTD2	Europe	KISEMHPAL	88.15%	38
	ORF9b protein		KVYPIILRL	80.84%	20
			GPKVYPIIL	60.10%	15
			RLVDPQIQL	55.65%	14
		North America	KISEMHPAL	85.77%	38
			KVYPIILRL	77.80%	20
			RLVDPQIQL	55.50%	15
			GPKVYPIIL	51.20%	14
		South Asia	KVYPIILRL	76.77%	20
			KISEMHPAL	65.26%	38
			GPKVYPIIL	32.99%	14
			RLVDPQIQL	31.48%	15
		North East Asia	KVYPIILRL	81.77%	20
			KISEMHPAL	81.09%	38
			RLVDPQIQL	64.31%	15
			SLEDKAFQL	37.92%	13

is homologous to template peptide GILGFVFTL (PDB: 2vlr), which is a linear peptidic epitope from matrix protein 1 of influenza A virus as recorded in IEDB and shows 61 peptides in peptide antigen family of template 2vlr across 34 organisms.

Peptide set from antigen P0DTD2 has not shown any hit peptide for any of the four target populations.

The hit peptide antigen SIIAYTMSL from P0DTC2 matches the profile of the homologous antigen family on positions 2, 4, 5, 8 and 9 (Fig. 3). The homologous peptide antigens prefers the nonpolar residues on second and fourth position (Met, Ile, Leu and Gly, Ala respectively) and the second position of the hit peptide is nonpolar residue Ile forming five VDW interactions with residues Tyr99, Val67, Met45, Tyr7, Phe9 and two hydrogen bonds with residues Lys 66, Glu63 on MHC molecule; fourth position of hit peptide is nonpolar residue Ala forming hydrogen bond with residue Gln52 in TCR. Position 5 of homologous peptide antigens prefers the aromatic residues (Phe, Tyr and Trp) and fifth position of hit peptide is aromatic residue Tyr forming strong VDW interaction with residue Leu156 on MHC molecule. Additionally position 8 of homologous peptide antigens prefers the polar residues (Ser, Thr and Asp) and Ser at position 8 in hit peptide forms VDW interaction with residue Thr73 and two hydrogen bonds with residues Trp147, Lys146 on MHC molecule and one hydrogen bond with residue Asp32 in TCR. Position 9 of homologous peptide

antigens prefers the nonpolar residues (Leu, Ile) and Leu at position 9 in hit peptide forms three VDW interactions with residue Leu81, Ile124, Trp147 and three hydrogen bonds with residue Asp77, Tyr84, Thr143 on MHC molecule.

Furthermore, the hit peptide antigen SPRWYFYYL from P0DTC9 relates the profile of the homologous antigen family on positions 2, 5, 7 and 9 (Fig. 4). Position 2 of homologous peptide antigens prefers the nonpolar residues (Ile, Leu, Met) and second position in the hit peptide is nonpolar residue Pro forming five VDW interactions with residue Tyr99, Val67, Met45, Tyr7, Phe9 and two hydrogen bonds with residue Lys66, Glu63 on MHC molecule. Position 5 and 7 of homologous peptide antigens prefers the aromatic residues (Phe, Tyr); fifth and seventh position of hit peptide are also aromatic residue Tyr forming strong VDW interaction with residue Leu156 and residue Leu156, Val152, Tyr166, Trp147 on MHC molecule respectively. Additionally position 9 of homologous peptide antigens prefers the nonpolar residues (Leu, Ile, Val, Met) and position 9 in the hit peptide is nonpolar residue Leu forming three VDW interactions with residue Leu81, Ile124, Trp147 and three hydrogen bonds with residue Asp77, Tyr84, Thr143 on MHC molecule.

Therefore, these two peptides can be considered as potential vaccine candidates and can be capable of evoking significant immune response. Further in-vivo/in-vitro



Fig. 1 PAComplex server showing pMHC-TCR interactions and homologous peptide for antigen P0DTC2

assessment should facilitate the effectiveness, development of polytopic vaccines and immune modulatory effects of the predicted peptides.

Conclusions

The world is in the midst of a COVID-19 pandemic. Vaccines can prevent infectious diseases and save millions of lives each year. Vaccines work by training and preparing the body's natural defences, the immune system, to recognize and fight off the viruses and bacteria they target. If the body is exposed to those disease-causing germs later, the body is immediately ready to destroy them, preventing illness. In recent years, peptide based vaccines have emerge as very convenient and crucial protection against infectious diseases. Immunoinformatics is a branch of bioinformatics that involves application of computational algorithms to analyse immunological data and problems. Advances in the field of immunoinformatics have led the development and widely distribution of hundreds of new vaccine design algorithms for exploration of proteomics. Prediction and analysis of antigenic peptides recognized by T helper and cytotoxic T lymphocytes from protein repertoire of pathogen followed by refined focus on the resulting set of peptides is central to modern vaccine development. The development of an effective and affordable vaccine against COVID-19 is the necessity of the hour for global public health. The present study involves application of various available bioinformatics tools for prediction of promising vaccine candidates by comprehensive mining of the proteome of SARS-COV-2. The pMHC-TCR interaction analysis in-silico demonstrated that the predicted peptides show homology to well-known potential antigens. Therefore, the present work is a very prominent strategy for rational antigen identification with further in-vivo/in-vitro experimentation required to emphasize the importance of the epitopes.



Fig. 2 PAComplex server showing pMHC-TCR interactions and homologous peptide for antigen P0DTC9



Fig.3 Frequency logo for the peptide antigen family of homologous template peptide loga (GILGFVFTL) of top hit peptide (SIIAYT-MSL)



Fig. 4 Frequency logo for the peptide antigen family of homologous template peptide 2vlr (GILGFVFTL) of top hit peptide (SPRW-YFYYL)

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Data Availability Data are available on request to the corresponding author.

Declarations

Conflict of interest Authors declare that they have no conflict of interest.

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