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In Silico Prediction of T-Cell and B-Cell Epitope in Mycobacterium Tuberculosis strain of H37Ra

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ABSTRACT: Mycobacterium tuberculosis leads the top cause of death, still in 2021 reported by the World Health Organization (WHO). In the near days, chemotherapy also becomes ineffective due to the development of resistance. As the infection spreads through droplets and airborne, it is necessary to prevent it through herd immunity through vaccinations. Since 1921, BCG is the only vaccine for tuberculosis, the BCG vaccine may be effective in children, but it is not protective even in younger adults. Since MDR-TB has been growing at a high rate, the treatment is becoming ineffective due to its wide range of resistance. The strains could also not respond to the immune system in TB individuals with lower immunity. Hence, new strategies and techniques have been created for the development of new vaccines. The virulent protein Ag85B from the strain H37Ra was selected as a vaccine candidate for the development of an effective vaccine. The protein sequence was retrieved from NCBI Genbank and the sequence was subjected for its antigenicity and allergenicity by in-silico software tools. The sequence was then predicted for its major histocompatibility complex against the human allele HLA-A*01:01 by the ProPred analyzing tool. The B-cell and T- cell epitope was then identified for probable antigens. Hence to conclude that Ag85B was found to be strong structural, desirable physiochemical and potential immunological attributes for the development of remarkable humoral and cellular immune response. Henceforth, Antigen 85B should be a potential lead candidate for in vitro and in vivo evaluations against Mtb.

Keywords: Mycobacterium tuberculosis, T-Cell, B-Cell, Vaccine, Histocompatibility, Epitope, Ag85B.

INTRODUCTION

Tuberculosis is a highly contagious infection that spreads globally, and the World Health Organization rated it as the leading cause of mortality from a single infectious agent (Andongma *et al.,* 2023). It was estimated that in 2019, the number of death cases reached 1.4 million, and that in 2021, the number of deaths was estimated to be 1.6 million, with 10.6 million new cases (Andongma *et al.,* 2023; Moodley *et al.,* 2022). Recently, the treatment that has been used is chemotherapy, which is a combination of antimicrobial medications. Chemotherapy can often be unsuccessful owing to the development of multi-drug resistance; hence, prevention of tuberculosis is better than treatment (Mitchison and Davies 2012). Mycobacterium TB in its latent stage affects the host and may become active at any time. The development of various treatments may also turn the host pathogens resistant to most of the treatments (Cohen *et al.,* 2019).

Vaccinations have been the most reliable prevention method for most infections; currently, the BCG vaccine for tuberculosis is effective in children, since its discovery in 1921. The BCG vaccine is unreliable in adults, particularly in immune-suppressed and immune-compromised patients (Gideon and Flynn 2011). They also lack the ability to develop cell mediated immune response that poses a low cure rate and it costs high for the treatment. The major threat also the human life faces is its co-infection with HIV, statistically, one-third of 34 million HIV infected humans were co-infected with Mtb (Shiraz *et al.,* 2021). Hence a new vaccine strategy is necessary to develop. Currently, 16 new vaccines are in different phases of clinical trials. Due to the time-consuming and labor-intensive nature of laboratory research on vaccine development, vaccine candidates can be developed by in-silico procedures. (Moodley *et al*., 2022).

The preparation of attenuated vaccines is an extensive process and the live vaccines have an increased risk of toxicity. The new era vaccine consists of B and T cell proteins that activate the immune response. The subunit vaccine depends on the T-cell complex that recognizes the cellular immune response (Ortega-Tirado *et al*., 2020). Effective subunit vaccine designs and selection should be based on the highest antigenic protein regions and the selection of more than one antigen at different stages of pathogens (Shiraz *et al.,* 2021).

The understanding of M.tb pathogenicity is still complex, due to its complex life cycle and spreading through aerosols released by the infected individuals.

The bacteria take advantage of innate immunity and persist in the host for a longer duration, 90% of infected individuals remain asymptomatic and only 10% of latent TB leads to symptomatic active TB. Since, the pathogen is lacking the common virulence factors such as flagella, toxins or capsules to invade the host, they usually rely on the host macrophages itself to cross the mucosal barrier to get avoid from phagocytosis. Firstly, the organism might weaken the endocytic pathway and induces innate immune system for their transmigration to the lung parenchyma cells, and then it induces the adaptive responses for its containment and start its action (Sharma and Sharma 2022).

The activation of macrophages' microbicidal pathway that activates CD4+ T-lymphocytes by antigenic epitope on antigen-presenting cells, MHC class II molecules leads to the release of pro-inflammatory cytokines. However, the CD4+ cells were not enough in clearing the latent Mtb, it was also necessary to activate CD8+ cells (Sharma and Sharma 2022).

The Mtb clearance is host dependent on the activation of macrophages' microbicidal pathway, antigen-presenting cells, activation of T lymphocytes, and release of cytokines. About 63% of antigenic epitopes were presented by MHC class II molecules and activated the CD4 T- lymphocytes. These lymphocytes secrete pro-inflammatory cells that activate the host's defense mechanism. The three isoenzymes of antigen 85 are (Ag85A, -B, and -C) are the most abundantly secreted proteins responsible for the outermost layer of the bacteria, that triggers the potent immune response and binds to fibronectin, due to their immunogenicity, Ag 85 A and B can also be considered as potential new vaccine candidate (Backus *et al.,* 2014).

The tubercular antigen is different from latently infected individuals to active TB cases, the immune-dominant antigen such as Ag85, ESAT-6, and CFP-10 can be selected as the potential vaccine candidate because of their expression in the early stage of infection (Sharma and Sharma 2022). The present study would investigate the suitable antigenic protein for the epitope prediction of the Mycobacterium tuberculosis H37Ra strain.

MATERIAL AND METHODS

The protein sequence of Ag85B of Mycobacterium tuberculosis was retrieved from NCBI Gen bank and the accession number is AAF13448.1 and consists of 283 amino acids. The Protein was then tested for its homology by the tool BLAST.

Sequence analysis. The sequence was then identified for its isoelectric point by Gene Runner, the comprehensive sequence analysis utility that features an amino acid table a codon frequency analyzer function of the isoelectric point, and its molecular weight. Restriction analysis is essential to develop the structure of protein, for mapping and sequencing. The tool NEB cutter finds the large, non-overlapping open reading frames using the *E. coli* genetic code and the sites for all type II restriction enzymes that cut the sequences.

Prediction of Antigenicity and Allegenicity. The sequence was then analyzed for its antigenic property and allergenicity by ANTIGEN pro (http://scratch.proteomics.ics.uci. edu/) and Aller TOP 2.0 (https://ddg-pharm fac.net/AllergenFP/), for the further analysis of MHC class I and Class II molecules by Propred and IEDB software for the allele (HLA-A*01:01) by SVM and ANN method with their IC 50 values.

Prediction of B-cell epitope. The prediction of B-cell epitope by BC Pred according to the hydro flexi surface polar properties with the flexi threshold value of 2, Hydrophilicity threshold of 1.9, exposed surface threshold value of 2.3, and polarity threshold value of 1.8 to identify the epitopes antigenicity, the individual epitope was found by Vaxigen 2.0.

Prediction of T-cell epitope. The prediction of T-cell epitope was done by CTL Pred which uses the 5 epitopes to find the perfect antigen that can be used as the vaccine candidate. Beta-turn prediction and their hydrophilicity nature by Kolaskar and Tongaonkar, Chou & Fasman beta-turn prediction tool and by Parker hydrophilicity prediction tool.

RESULTS AND DISCUSSION

The Mycobacterium tuberculosis is the most common cause of tuberculosis and among all the virulence factors, the Ag85 complex is predominant. The protein Ag85 complex is a family of 3 proteins such as Ag85A, Ag85B, and Ag85C, which possess the mycolyl-transferase action that is bound to the arabinoglycoside of the cell wall. These proteins are also called fibronectin-binding proteins that had severe virulence action on human cells (Kremer *et al.,* 2002), hence the targeted protein for novel vaccine development is Ag85B. The protein sequence of Antigen 85B precursor in Mycobacterium tuberculosis H37Ra retrieved from NCBI and the Genbank Id is AAF13448.1.

>AAF13448.1 antigen 85-B precursor, partial [Mycobacterium tuberculosis H37Ra]
FSRPGLPVEYLQVPSPSMGRDIKVQFQSGGNNSPAVY
LLDGLRAQDDYNGWDINTPAFEWYYQSGLSIVMPVGG
QSSFYSDWYSPACGKAGCQTYKWETFLTSELPQWLSA
NRAVKPTGSAAIGLSMAGSSAMILAAYHPQQFIYAGS
LSALLDPSQGMGPSLIGLAMGDAGGYKAADMWGPSSD
PAWERNDPTQQIPKLVANNTRLWVYCGNGTPNELGGA
NIPAEFLENFVRSSNLKFQDAYNAAGGHNAVFNFPPN
GTHSWEYWGAQLNAMKGDLQSSLG

Prediction of restriction sites. The restriction fragments of the given protein sequence were analyzed by the NEB cutter tool to find 3 enzymes flank the regions 52-868 within 200bp. The results also showed a GC content of about 64% and AT of about 36% (Fig. 1).

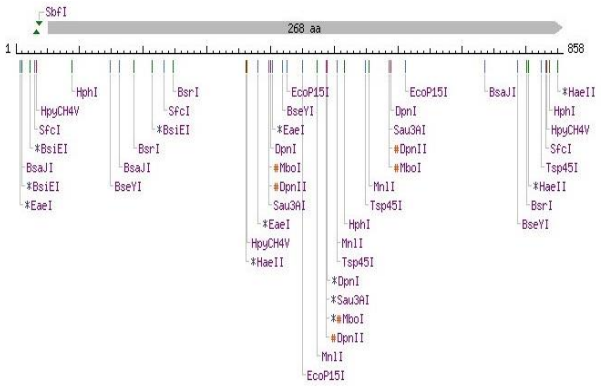

Fig. 1. Restriction sites mapping.

Prediction of antigenicity and Allergenicity. The antigenic property of selected vaccine candidate Ag85 was predicted by the tool ANTIGEN pro and obtained predicted antigenicity was 0.817334. The predicted antigenicity was similar to the results obtained by Moodley *et al.* (2022) for the MTB protein sequence of PE_PGRS17 with a score value of 0.8724. The allergic property of the protein for the suitable vaccine candidate by the software Allertop reveals that protein Ag85B is a non-allergen protein.

The nature of the protein and its iso-electric point was analyzed using the software GENE RUNNER and it is found that the iso-electric point is 5.41. The molecular weight of a protein sequence was analyzed by an online tool called Sequence manipulation suite, the results showed for 268 amino acid residues were 22.83kDa.

Prediction of B-cell. B-cell epitope prediction was done by BC Pred and it identifies about 5 epitopes with common properties of flexibility, Polarity, Hydrophilicity and Exposed surface. The epitope prediction also done by IEDB analysis resource showed 5 predicted peptides with an average score value of about 0.4 (Fig. 2a.), the beta-turn prediction showed the average of 1.063 by Chou & Fasman beta-turn prediction tool (Fig 2b), the antigenicity was done by Kolaskar and Tongaonkar showed the average of 1.015 (Fig 2c.) with the scale of Q and the hydrophilic property was predicted by Parker hydrophilicity prediction showed the value of 1.534 (Fig. 2d). The antigen probability was identified by the Vaxigen score for all 5 epitopes. The sequence NAAGGHN showed the highest vaxigen score value of 2.4816 with probable antigenicity (Table 1).

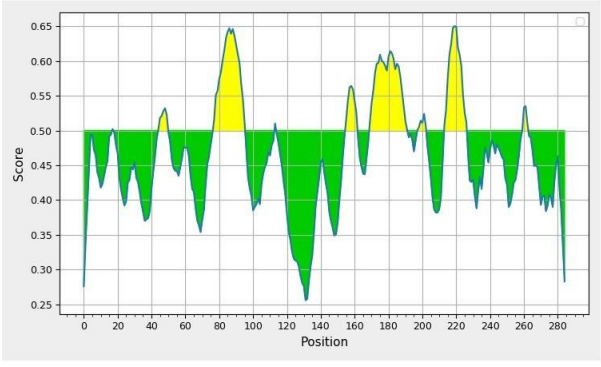

Fig. 2a. Bepipred Linear Epitope Prediction 2.0.

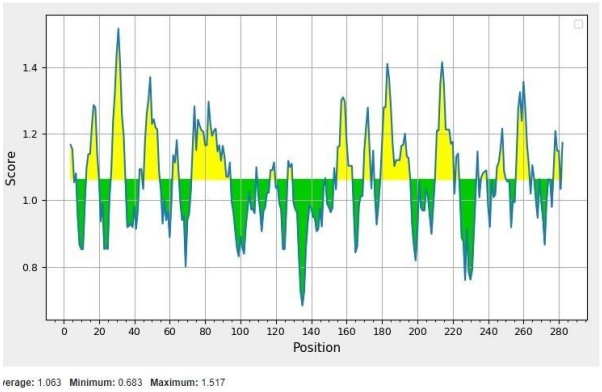

Fig. 2b. Chou & Fasman Beta-turn Prediction.

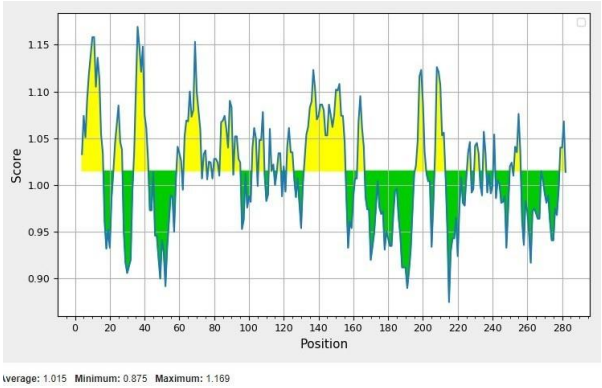

Fig. 2c. Antigenic property by Kolaskar and Tongaonkar Antigenicity

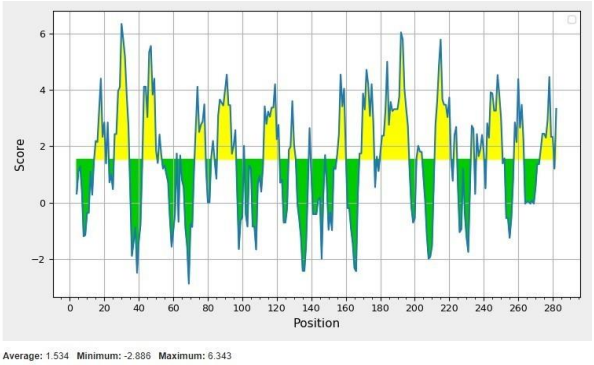

Fig. 2d. Parker Hydrophilicity results.

Table 1: Vaxigen score for all the 5 epitopes.

Common Properties	Sequence	Vaxigen Score	Antigen Probability
Hydrophilicity and Flexibility	QSGGNNSPA	1.640	Probable Antigen
Hydrophilicity and Flexibility	GPSSDPA	0.295	Non-Antigen
Hydrophilicity, Flexibility, Polarity and exposed surface	ERNDPTQQ	1.392	Probable Antigen
Hydrophilicity and Flexibility	CGNGTPNEL	1.7108	Probable Antigen
Hydrophilicity and Exposed surface	**NAAGGHN**	**2.4816**	**Probable Antigen**

Prediction of T-cell epitope. T cell epitope prediction was done by CTL Pred that showed the top 5 epitopes on the protein sequence with a high score of 1.254 on

the 144th position of sequence IYAGSLSAL by SVM tool (Table 2) and with a cut-off score of 0.34 and by ANN tool the obtained score value of 1st rank is 0.990 on 140th position with a cut-off score of 0.51of sequence PQQFIYAGS (Table 3). Among all the 5 predicted epitopes, the sequence RPGLPVEYL was predicted as the probable antigen with a score of 0.5177 of the threshold value of 0.4.

Table 2: Prediction of T-cell epitopes by CTL Pred (only SVM).

Peptide Rank	Position	Sequence	Score	Prediction
1	144	**IYAGSLSAL**	1.254	Epitope
2	108	WLSANRAVK	1.128	Epitope
3	3	RPGLPVEYL	1.018	Epitope
4	158	**GMGPSLIGL**	1.001	Epitope
5	224	IPAEFLENF	1.000	Epitope

Table 3: Prediction of T-cell epitopes by CTL Pred (only ANN).

Peptide Rank	Position	Sequence	Score	Prediction
1	140	**PQQFIYAGS**	0.990	Epitope
2	158	**GMGPSLIGL**	0.990	Epitope
3	177	ADMWGPSSD	0.990	Epitope
4	41	GLRAQDDYN	0.960	Epitope
5	61	YYQSGLSIV	0.950	Epitope

Prediction of MHC molecules by IEDB database and MHC Pred. The major histocompatibility complex (MHC) class I and MHC class II were analyzed by IEDB software by SMM and ANN methods. For MHC class I the results showed for Human allele (HLA-A*01:01) predicted as high efficiency. The MHC class II molecules can be predicted using the tool SMM_align 1.1 showed the antigen has a very low IC 50 and it also showed that Ag85B is a good binder molecule.

The MHC also predicted by MHC Pred for the two sequences such as IYAGSLSAL and GMGPSLIGL that scored high in CTL pred and BC Pred software against four human alleles such as A0101, DRB0101, DRB0401, and DRB0701. Of all four, DRB0401 showed the high score and predicted IC50 of 5.878 and 5.786 for the two sequences respectfully (Table 4). The sequences also showed they are digested by only two enzymes by Peptide cutter (Fig. 1).

Table 4: Selected sequences against the alleles and its IC50.

Sequence	Allele	Predicted IC50 (M)	Predicted IC50 (nM)	Proteasomal activity by Peptide cutter
IYAGSLSAL	A0101	-	-	Digested by Pepsin, chymotrypsin and Thermolysin
	DRB0101	**7.627**	**23.60**	
	DRB0401	5.878	1324.34	
	DRB0701	5.472	3372.87	
GMGPSLIGL	A0101	-	-	Digested by Pepsin, chymotrypsin and Thermolysin
	DRB0101	**6.664**	**216.77**	
	DRB0401	5.768	1706.08	
	DRB0701	5.972	1066.60	

The increased number of multidrug-resistant tuberculosis and the development of tuberculosis association with other diseases like HIV-positive patients emphasize the necessity to find new antitubercular targets and drugs (Favrot *et al.*, 2014). Antigen 85 (Ag85) received much attention from scientists to develop a vaccine against Mtb rather than the other immunogens. Because the Ag85 allows the bacteria to escape from the host immune response by foiling the formation of phagolysosomes. Since, phagolysosomes are plays an essential role in innate immune response for eradication of infection (Babaki *et al*., 2017).

In our present investigation, we found that the antigenicity score of selected vaccine candidate Ag85 was 0.82. Similarly, Sharma *et al*. (2021) reported that the antigenicity of the constructed vaccine against Mtb was predicted as 0.95 through VaxiJen and AntigenPro web servers. The highly antigenic properties of the selected targets in the Insilco parameters are crucial in the development of vaccines before the beginning of the wet lab studies. Since, the antigenicity provided the primary evidence of, how the body's innate and acquired (B cell and T cell receptors) immune responses will react against the antigens (Fishman *et al.,* 2015). Our Insilco prediction showed the antigenicity was more than the threshold value (0.5) and considered a suitable target for vaccine development.

The molecular weight of the selected antigen showed 22.83 kDa with 268 numbers of amino acid residues with an iso-electric point of 5.41. In contrast with our study, Bibi *et al.* (2021) reported that the selected antigen such as Rv2608, Rv2684, Rv3804c, and Rv0125 against Mtb showed a molecular weight of more than 31.9kDa with the iso-electric point of 4.28 score. Since, the peptide vaccine with higher molecular weight showed an efficient function in the alleviation of the disease (Sharma *et al*., 2021).

In our present investigation, the selected vaccine antigen Ag85 showed higher affinity against MHC class I molecules with low IC50 value. It's essential to find out the binding efficiency of vaccine candidates into the MHC molecules. These peptides are generated by proteolysis of endogenously synthesized proteins in the cytosol, loaded onto MHC-I molecules, and presented on the cell surface for surveillance by CD8(+) T cells. MHC-I restricted processing and presentation alerts the immune system to any infectious unfolding intracellularly and provides potential targets for a cytotoxic T-cell response. Therefore, therapeutic vaccines based on MHC-I presented peptide epitopes could, theoretically, induce CD8(+) T cell responses that have tangible clinical impacts on disease eradication (Comber and Philip 2014).

Leddy *et al.* (2023) has also found that MHC class I antigen have been identified as the potential vaccine candidate, due to its immunogenic responses against the virulent proteins ESx, as they are the source of immunodominant CD8(+) T cells antigens and they are protective against Mtb infection.

Our present findings on B-cell epitope prediction through ABC pred showed about 9 epitopes were explored flexi threshold value with high antigenicity and selected for the vaccine development. On the other hand, 4 numbers of T cell epitopes were selected through CTL pred. Likewise, Bibi *et al.* (2021), selected the four numbers of B-cell epitope with high antigenic, non-allergenic, and non-toxic, a total of vaccine construction. Both the B-cells and T- cells epitopes ideally can be used to develop vaccines against Mtb and may be reliable for inducing both humoral and cell-mediated immunity (Dubey *et al.*, 2018).

The other study Al Tbeishat (2022) reported that overall 8 B cell epitopes and 17 T cell epitopes were selected after the exclusion criteria for the vaccine development against tuberculosis. The importance of the selection of B and d Cell epitopes in vaccine development is very crucial, since both components are playing a major role in the immune system in fighting against pathogens (Saha and Raghava 2006). Initially, the primary immune response against infections was mediated by the elevated level of the IgM antibodies after a lag period of 5–7 seven days of antigen exposure.

The online tool MHC Pred showed specificity for the identification of vaccine candidates against the suitable allele. Interestingly, the vaccine epitope by the software had 99.9% of population coverage. The results of the present study were correlated with Medha *et al.* (2022) reported peptide-based vaccines against PE/PPE proteins of Mtb.

Continuation with that, the secondary immune response is developed by the increased proliferation of B-cell as well as increased expression of IgM, IgG1 + IgG2 and IgG + IgM antibodies (Sharma *et al*., 2021; Jung *et al*., 2011). On the other hand, previous studies revealed that T cell epitopes can potentially enhance the production of antigen-specific IgG antibodies and enhance the antibody response. Also produced the T helper 1 (Th1)-type response and promoted CD4+ T cell proliferation and was strongly involved in the cell-mediated immune response against any kind of infection (Fan *et al*., 2021; Choi *et al*., 2015).

CONCLUSIONS

The Antigen 85B of Mycobacterium tuberculosis is found to be the most predominant protein in virulent action. Thereby, the protein was selected as an antigenic molecule as a vaccine candidate. The protein was analyzed by various online tools to identify its antigenicity and Allergenicity, revealing that it is non-allergic for the allele HLA-A*01:01. The T cell and B cell epitopes were also predicted by CTL Pred and BC Pred software to obtain the sequence RPGLPVEYL has probable antigenic property with the score value of 0.5177 among 285 amino acid residues. The MHC Pred software concluded that low IC50 was found in the allele DRB*0101. Overall, the present finding is concluding that the selected Antigen 85B showed strong structural, desirable physiochemical and potential immunological attributes that can lead to the development of remarkable humoral and cellular immune responses. Henceforth, Antigen 85B should be a potential lead candidate for in vitro and in vivo evaluations against Mtb.

FUTURE SCOPE

The promising present finding is the selected Antigen 85B showed a strong structural, desirable physiochemical and potential immunological attribute that can lead to the development of remarkable humoral and cellular immune response.

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Conflict of Interest. None.

REFERENCES

Al Tbeishat, H. (2022). Novel In Silico mRNA vaccine design exploiting proteins of M. tuberculosis that modulates host immune responses by inducing epigenetic modifications. *Scientific Reports, 12*(1), 1-19.

Andongma, B. T., Huang, Y., Chen, F., Tang, Q., Yang, M., Chou, S. H. & He, J. (2023). In silico design of a promiscuous chimeric multi-epitope vaccine against *Mycobacterium tuberculosis*. *Computational and Structural Biotechnology Journal.*

Babaki, M. K. Z., Soleimanpour, S. & Rezaee, S. A. (2017). Antigen 85 complex as a powerful *Mycobacterium tuberculosis* immunogene: Biology, immune-pathogenicity, applications in diagnosis, and vaccine design. *Microbial pathogenesis, 112*, 20-29.

Backus, K. M., Dolan, M. A., Barry, C. S., Joe, M., McPhie, P., Boshoff, H. I. Barry, C. E. (2014). The three *Mycobacterium tuberculosis* antigen 85 isoforms have unique substrates and activities determined by non-active site regions. *Journal of Biological Chemistry, 289*(36), 25041-25053.

Bibi, S., Ullah, I., Zhu, B., Adnan, M., Liaqat, R., Kong, W. B. & Niu, S. (2021). In silico analysis of epitope-based vaccine candidate against tuberculosis using reverse vaccinology. *Scientific reports, 11*(1), 1249.

Choi, H. G., Kim, W. S., Back, Y. W., Kim, H., Kwon, K. W., Kim, J. S. & Kim, H. J. (2015). *Mycobacterium tuberculosis* RpfE promotes simultaneous Th1-and Th17-type T-cell immunity via TLR4-dependent maturation of dendritic cells. *European journal of immunology, 45*(7), 1957-1971.

Cohen, A., Mathiasen, V. D., Schön, T. & Wejse, C. (2019). The global prevalence of latent tuberculosis: a systematic review and meta-analysis. *European Respiratory Journal, 54(*3).

Comber, J. D. & Philip, R. (2014). MHC class I antigen presentation and implications for developing a new generation of therapeutic vaccines. *Therapeutic advances in vaccines, 2*(3), 77-89.

Dubey, K. K., Luke, G. A., Knox, C., Kumar, P., Pletschke, B. I., Singh, P. K. & Shukla, P. (2018). Vaccine and antibody production in plants: developments and computational tools. *Briefings in functional genomics, 17*(5), 295-307.

Fan, X., Li, X., Wan, K., Zhao, X., Deng, Y., Chen, Z. & Liu, H. (2021). Construction and immunogenicity of a T cell epitope-based subunit vaccine candidate against *Mycobacterium tuberculosis*. *Vaccine, 39*(47), 6860-6865.

Favrot, L., Lajiness, D. H. & Ronning, D. R. (2014). Inactivation of the *Mycobacterium tuberculosis* antigen 85 complex by covalent, allosteric inhibitors.

Journal of Biological Chemistry, 289(36), 25031-25040.

Fishman, J. M., Wiles, K. & Wood, K. J. (2015). The acquired immune system response to biomaterials, including both naturally occurring and synthetic biomaterials. In Host response to biomaterials (pp. 151-187). Academic Press.

Gideon, H. P. & Flynn, J. L. (2011). Latent tuberculosis: what the host "sees"? *Immunologic research, 50, 202*-212.

Jung, I. D., Jeong, S. K., Lee, C. M., Noh, K. T., Heo, D. R., Shin, Y. K. & Park, Y. M. (2011). Enhanced Efficacy of Therapeutic Cancer Vaccines Produced by Co-Treatment with *Mycobacterium tuberculosis* Heparin-Binding Hemagglutinin, a Novel TLR4 AgonistM. tuberculosis HBHA for Cancer Vaccines. *Cancer Research, 71*(8), 2858-2870.

Leddy, O., White, F. M. & Bryson, B. D. (2023). Immunopeptidomics reveals determinants of *Mycobacterium tuberculosis* antigen presentation on MHC class I. *Elife, 12*, e84070.

Kremer, L., Maughan, W. N., Wilson, R. A., Dover, L. G. & Besra, G. S. (2002). The *M. tuberculosis* antigen 85 complex and mycolyltransferase activity. *Letters in applied microbiology, 34*(4), 233-237.

Medha, M. & Roy, A. (2022). Microgravity: New aspect for breast cancer treatment, a review. *Acta Astronautica, 190*, 62-73.

Mitchison, D. A. V. I. E. S. G. & Davies, G. (2012). The chemotherapy of tuberculosis: past, present and future [State of the art]. *The international journal of tuberculosis and lung disease, 16*(6), 724-732.

Moodley, A., Fatoba, A., Okpeku, M., Emmanuel Chiliza, T., Blessing Cedric Simelane, M. & Pooe, O. J. (2022). Reverse vaccinology approach to design a multi-epitope vaccine construct based on the *Mycobacterium tuberculosis* biomarker PE_PGRS17. *Immunologic Research, 70*(4), 501-517.

Ortega-tirado, D., Arvizu-fores, A. A. & Velazquez, C. (2020). Funding: This work was supported by Consejo Nacional de Ciencia y Tecnología. *Expert Rev Vaccines.*

Saha, S. & Raghava, G. P. S. (2006). Prediction of continuous B-cell epitopes in an antigen using recurrent neural network. Proteins: Structure, Function, and Bioinformatics, *65*(1), 40-48.

Sharma, S. & Sharma, M. (2022). Design of a peptide-based vaccine from late stage specific immunogenic cross-reactive antigens of PE/PPE proteins of *Mycobacterium tuberculosis*. *European Journal of Pharmaceutical Sciences, 168*, 106051.

Sharma, R., Rajput, V. S., Jamal, S., Grover, A. & Grover, S. (2021). An immunoinformatics approach to design a multi-epitope vaccine against Mycobacterium tuberculosis exploiting secreted exosome proteins. *Scientific Reports, 11*(1), 13836.

Shiraz, M., Lata, S., Kumar, P., Shankar, U. N. & Akif, M. (2021). Immunoinformatics analysis of antigenic epitopes and designing of a multi-epitope peptide vaccine from putative nitro-reductases of *Mycobacterium tuberculosis* DosR. Infection, *Genetics and Evolution, 94*, 105017.

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