**Biotechnology International 3 (2/3): 29-32, September 2010**
**ISSN 0974-1453**

**Published by Biotechnology Society Online** www.bts.org.in

# Prediction of MHC-I binding epitopes in a gene fragment encoding 183 amino acids of *Mycobacterium bovis* Bacillus Calmette-Guerin (BCG) strain

**Ramesh.D[1*]., Sadeesh .E.M[1]., R . Deb[2]., B. Sailo[3]., V. K. Saxena[2]., V.G Joshi [2]and Yosef .D[2]**

Indian Veterinary Research Institute Izatnagar- 243122 U.P India, [1]PhD Scholar Division of Animal Biochemistry, [2]PhD Scholar Division of Animal Biotechnology,[3]PhD Scholar Division of Veterinary Public Health
* Corresponding author- drrameshd@gmail.com

**Summary:** In this study, *in silico* prediction of nanomeric epitopes of *Mycobacterium bovis* BCG str. Tokyo 172 was explored, and analyzed for CD8+ T cell binding ability based on their predicted peptide scores for their respective MHC-I specific alleles

**Keywords: Mycobacterium bovis, MHC-I**

## Introduction

Tuberculosis continues to affect about 30% of the world's population, particularly in developing countries, Bacillus Calmette-Guerin (BCG) vaccination efficacy is controversial, and seems to fail to protect adults against pulmonary form of tuberculosis (Bloom and Mckinney, 1999). Targeting the *Mycobacterium tuberculosis* using subunit vaccine with virulence proteins by assembling their minimal CD8+ and CD4+ T cell epitopes might be effective in inducing immune responses. Recently the complete sequence of *Mycobacterium bovis* BCG Tokyo 172 genome was determined and compared with BCG Pasteur and other *M. tuberculosis* complex (Seki *et al*., 2009).The present study aimed towards the prediction of CD8+ antigenic peptides (9- mers) from the portion of *Mycobacterium bovis* Bacillus Calmette-Guerin (BCG) strain Tokyo 172 accession AP010918.1 that binding to class I MHC CD8+ molecules. This prediction will be useful in designing new peptide molecules for antigen-based vaccine design.

## Materials and Methods

The hypothetical protein of *Mycobacterium bovis* BCG str Tokyo 172 GenBank accession AP010918.1 (183 amino acid) was used in this study. The bioinformatics tool, ProPred1 (Singh and Raghava, 2003) was used to predict MHC class I binding peptides

(Cytotoxic T-Lymphocytes CTL epitopes) for 47 alleles by adjusting the threshold score of 4% in presence of Proteosome and Immuno Proteosome Filter.

**Results and Discussion**

In this study, *in silico* prediction of nanomeric epitopes of *Mycobacterium bovis* BCG str. Tokyo 172 was explored, and analyzed for CD8+ T cell binding ability based on their predicted peptide scores for their respective MHC-I specific alleles. All possible overlapping 9-mer peptides were generated for a given antigen sequence. The scores of this 9-mer peptide were calculated using quantitative matrix of selected MHC alleles. All peptides having score greater than selected threshold score (4%) were assigned as predicted binders for selected MHC-I alleles as indicated by others (Bhasin, 2003; Somvanshi P *et al.,* 2008).

However among 47 alleles screened, top 26 antigenic CTL epitopes (25 for Human and 1 for Cattle MHC-I) has been selected (which were showing score > 50) depicted in the table .1. Out of 26 CTL epitopes were predicted, the peptides, MHC-Kd (4800), HLA-B*2705 (2000), HLA-B14 (1000) and HLA-B*5102 (880) are the top four scorer, with maximum MHC-I binding ability.

Bioinformatics tools had the potential to accelerate research into the design of vaccines and diagnostic tests by exploiting genome sequences. Moreover the predicted nanomeric epitopes can be evaluate of *in vivo* and *in vitro* studies confirmed further for their binding ability to MHC-I, their efficiency in inducing strong immune responses as potent vaccine candidates can be evaluated in future.

**Table 1. The predicted MHC-I epitopes using the ProPred1 for the *Mycobacterium bovis***

| **Alleles** | **Predicted MHC-I epitopes sequence** | **Amino acid position** | **Score** | **Predicted binder /non binder** |
|---|---|---|---|---|
| **HLA-A2** | RLYGVAYTL | 85 | 279.576 | Predicted Binder |
| **HLA-A*0201** | ILSVGEHSV | 50 | 118.238 | Predicted Binder |
| **HLA-A24** | IYLVGQMAL | 99 | 300.000 | Predicted Binder |
| **HLA-A68.1** | DVYRFLLRR | 74 | 600.000 | Predicted Binder |
| **HLA-A20**<br><br>**Cattle** | RKLKINTIL | 43 | 500.000 | Predicted Binder |

**BCG str. Tokyo 172 protein.**

| | | | | |
|---|---|---|---|---|
| **HLA-A2.1** | RLYGVAYTL | 85 | 124.600 | Predicted Binder |
| **HLA-B14** | WRLSRGESL | 148 | **1000.000** | Predicted Binder |
| **HLA-B*2705** | WRLSRGESL | 148 | **2000.000** | Predicted Binder |
| **HLA-B*3701** | VDSDFNALL | 126 | 200.000 | Predicted Binder |
| **HLA-B*3901** | NREDVYRFL | 71 | 90.000 | Predicted Binder |
| **HLA-B*5101** | LPGERKLKI | 39 | 629.200 | Predicted Binder |
| **HLA-B*5102** | LPGERKLKI | 39 | **880.000** | Predicted Binder |
| **HLA-B*5103** | KPDENREDV | 67 | 58.080 | Predicted Binder |
| **HLA-B*5201** | GGAPPALIV | 29 | 60.000 | Predicted Binder |
| **HLA-B*5301** | LPGERKLKI | 39 | 123.010 | Predicted Binder |
| **HLA-B*5401** | FNALLELGF | 130 | 176.330 | Predicted Binder |
| **HLA-B*51** | LPGERKLKI | 39 | 131.960 | Predicted Binder |
| **HLA-B60** | VELPGERKL | 37 | 352.000 | Predicted Binder |
| **HLA-B62** | LLRRNRRLY | 79 | 60.000 | Predicted Binder |
| **HLA-B7** | APPALIVEL | 31 | 240.000 | Predicted Binder |
| **HLA-B*0702** | HPRPGGAPP | 25 | 147.860 | Predicted Binder |
| **HLA-Cw*0301** | QNLQAFAHL | 157 | 100.000 | Predicted Binder |
| **HLA-Cw*0401** | IYLVGQMAL | 99 | 200.000 | Predicted Binder |
| **MHC-Db** | YTLDNVGDI | 91 | 600.000 | Predicted Binder |
| **MHC-Kd** | IYLVGQMAL | 99 | **4800.000** | Predicted Binder |
| **MHC-Kk** | LELGFRSSI | 134 | 500.000 | Predicted Binder |

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