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T-Cell Epitope Prediction of Dengue Virus (DENV) Non-Structural-1 (NS1) Gene Using TepiTool Prediction Tools

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Background: Dengue virus (DENV) remains a major public health concern in Southeast Asia and is transmitted by *Aedes* mosquitoes. DENV exists as four distinct serotypes, which complicates vaccine development. Although current vaccine strategies can elicit immune responses, they often fail to provide effective, balanced protection against all four serotypes and may pose safety concerns. In addition, traditional vaccine development approaches are time-consuming and costly. Therefore, this study applied a reverse vaccinology and immunoinformatics approach to identify potential T-cell epitopes from the dengue virus non-structural 1 (NS1) gene. This strategy offers a faster, more cost-effective method for identifying conserved and specific epitopes across diverse viral strains, thereby contributing to the development of a multi-epitope dengue vaccine.

Methods: NS1 gene sequences from all four dengue virus serotypes were retrieved from the NCBI Virus and GenBank databases. T-cell epitopes from the NS1 protein were predicted using the TepiTool platform. Epitope conservation analysis was subsequently performed using the IEDB database, and conserved regions were visualised using WebLogo version 3.0. In addition, BLASTp analysis was conducted to assess similarity between the predicted epitopes and human proteins.

Results: A total of 48 NS1 protein sequences from DENV-1 to DENV-4 were successfully retrieved from the databases. NS1 regions were extracted from dengue polyprotein sequences to ensure accurate analysis. The sequences were obtained from dengue-endemic Southeast Asian countries, including Malaysia, the Philippines, Vietnam, Thailand, Indonesia, and Myanmar. T-cell epitope prediction using TepiTool was performed across 12 MHC Class I and 6 MHC Class II alleles. Epitope selection was based on percentile rank, with lower values indicating stronger predicted binding affinity. Percentile ranks below 1.0 for MHC Class I and below 10.0 for MHC Class II were selected. Overall, eight conserved epitopes were identified, comprising six MHC Class I and two MHC Class II epitopes with 80 to 100 per cent sequence conservation across all serotypes.

Conclusion: This study demonstrates that immunoinformatics tools provide a rapid, safe, and effective framework for early-stage dengue vaccine design. The identified conserved T-cell epitopes from the NS1 protein are promising candidates for further experimental validation and the development of multi-epitope dengue vaccines.

Keywords: Dengue Virus, Reverse vaccinology, NS1 protein, T-cell Epitopes.

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