ENGINEERING OF A NANOMOLECULAR PROTEIN OF CHIKUNGUNYA VIRUS TOWARDS DEVELOPING A HIGH-THROUGHPUT ANTIVIRAL SCREENING PLATFORM

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Abstract

Chikungunya fever, a debilitating infection triggered by the Chikungunya virus, has always been perceived as a neglected tropical disease. Lack of approved prophylactic/therapeutic interventions for this disease propels the need to expedite the drug discovery process. Several screening approaches have been tried to identify potential antiviral molecules targeting the viral proteins specifically. However, these approaches target the whole virus, which has its associated constraints. Therefore, as an alternative approach, the present study was aimed at utilizing the enzymatic function of a specific viral protein to develop an in-house cell-free based high throughput antiviral screening platform

The capsid protein (CP) of chikungunya virus (CHIKV), is a structural and multifunctional protein with proteolytic activity, which was the selected viral target in the present study. The chikungunya virus capsid protein (CVCP) was expressed using prokaryotic host systems. To achieve this, viral RNA was isolated from an unlinked anonymous chikungunya virus real time PCR positive sample and cDNA was synthesized from the extracted RNA. CHIKV structural polyprotein (SP) gene was amplified using cDNA as template, cloned into vector, and transformed. Further, capsid gene was amplified using SP gene, the product was sub-cloned into expression vector, and transformed in a prokaryotic host system. Capsid gene was confirmed by molecular sequencing. Subsequently, capsid protein was expressed using standardized protocol, confirmed by western blot and purified to obtain in microgram quantities. This method may be useful to synthesize viral proteins in-house in bulk quantities, which otherwise are expensive to procure commercially. Further, these proteins may serve as nanomolecular targets to study drug-protein interactions towards development of high-throughput antiviral screening platforms.