Title: "Evaluation of potential interceptors targeting non-structural protein-2 of chikungunya virus"

ABSTRACT

Chikungunya virus (CHIKV) is a pathogen for chikungunya fever (CHIKF) that is characterized by maculopapular rashes, high-grade fever, stomach upsets, headache, myalgia, polyarthritis, and polyarthralgia. The disease may confer fatality in its severe form due to central nervous system invasion and ending up in encephalitis. There are no FDA approved therapeutic interventions available to tackle the disease. Herein, we tried to investigate the new inhibitors of CHIKV replication using various approaches. The first approach was to establish the new role of approved molecules as an antiviral against CHIKV. The multi-enzymatic and multi-functional non-structural protein-2 (nsP2) was targeted to develop its inhibitors. We adopted a systematic pathway of i*n- silico* virtual screening through docking (ParDOCK) and molecular dynamics simulations. The best molecules were subjected to physical interactions with recombinant nsP2 using biophysical techniques of surface plasmon resonance, small-angle x-ray scattering, and fluorescence-based assays.

The protease activity inhibition was monitored using screened molecules as inhibitors. Further, the molecules were subjected to plaque reduction and neutralization test (PRNT) to assess their virus inhibitory potential. This study resulted in establishing the anti-CHIKV role of novobiocin and telmisartan. Novobiocin displayed ameliorating effects in murine model of CHIKV. The second approach was using aqueous extracts of medicinal plants and herbs. The plants were shortlisted by *Ayurvedic* literature review and tested against the helicase and protease inhibition. The PRNT was used to evaluate their potential to inhibit the CHIKV in cell culture using Vero cells. The study gave three plants *Ocimum tenuiflorum*, *Terminalia chebula*, *Picrorhiza kurroa* that inhibited nsP2 enzymatic activity and plaques as well. The third approach was to evaluate the phytomolecules as nsP2 inhibitors. The molecules were screened by in-silico and biophysical methods. We present naringin that binds nsP2 with very high affinity.