Abstract

Dengue virus remains a major global health threat, yet no dengue-specific antiviral therapy has been approved. The development of rapid and scalable antiviral screening platforms is therefore essential to accelerate early-stage drug discovery. This study aimed to optimize and evaluate a recombinant DV-2 eGFP reporter virus system for high-throughput antiviral screening. DV-2 eGFP virus was rescued and amplified in BHK-21 cells, with viral replication monitored through GFP fluorescence. Fluorescence intensity peaked at Day 6 post-infection, corresponding to the highest infectious virus production. Infection conditions were further optimized across Huh-7, THP-1 DC-SIGN, and THP-1 TIM-1 cells, where Huh-7 cells demonstrated the strongest GFP signal and highest infection efficiency. The system was validated using Ribavirin and NITD008 as reference antiviral compounds. NITD008 showed stronger antiviral activity, with an EC_{50} of 0.57 μM, CC_{50} of 35.74 μM, and selectivity index of 63.09, whereas Ribavirin showed lower selectivity. Assay robustness was confirmed by a Z′-factor of 0.72, indicating excellent suitability for screening. Pilot screening of 40 compounds identified 6 candidates exceeding the 50% inhibition threshold. Overall, the optimized DV-2 eGFP reporter system provides a rapid, fluorescence-based platform for dengue antiviral screening, although further improvement of viral titer and reporter stability is recommended.

Keywords: Dengue virus, DV-2 eGFP, antiviral screening, reporter virus, high-throughput screening, Huh-7 cells