

Stable Dengue Virus Infectivity

The measurement of neutralizing and enhancing antibodies against Dengue virus (DENV) is commonly employed to assess the efficacy of potential Dengue vaccines and to determine the protective and pathogenic humoral response to natural infection. These applications can involve analysis of a large number of serum samples. In accordance, a standardized and stable detection system for DENV infection is required for reliable, accurate readouts following diverse incubation times and conditions used in experimental protocols.

The Dengue Reporter Virus

The Dengue Reporter Virus Particle (RVP) enables the rapid screening of human sera for Dengue neutralizing and enhancing antibodies. RVPs are replication-incompetent reporter viruses that maintain antigenic equivalence to each of the four serotypes of DENV. Pre-incubation of virions with test sera, typically for 1-2 h, is common in most neutralization and enhancement assays but requires the virus to maintain infectivity after the incubation period. The stability of live DENV and its ability to form plaques under such conditions can vary. The RVP is designed to permit objective and quantitative readouts of infectivity under all standard experimental conditions, including pre-incubation at varying temperatures and storage for prolonged periods of time. The ability to prepare and store large amounts of RVPs for repeated experimentation can reduce the variability of using different batches of reagents.

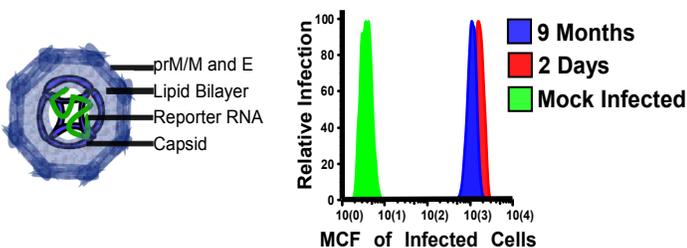


Figure 1. Dengue Reporter Virus Retains Infectivity Following Long-term Cryo-storage. RVPs were stored for the indicated times at -80°C before being thawed, mixed with Raji DC-SIGNR cells, and cultured for 48 hours. Infection was quantified by flow cytometry (Mean Cell Fluorescence of gated cells).

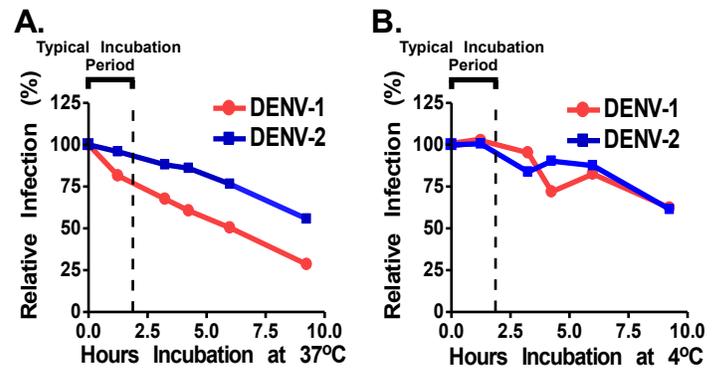


Figure 2. Dengue Reporter Virus Retains Strong Infectivity After Prolonged Incubations at Different Temperatures. RVPs specific for serotypes 1 (DENV-1) and 2 (DENV-2) were incubated for the indicated times at either 37°C (A) or 4°C (B), then mixed with Raji DC-SIGNR cells and cultured for 48 hours. Infection was quantified by flow cytometry.

Technical Description

RVPs have proven resilient in many testing environments. The stability of RVP particles has been tested in infection assays following cryopreservation for several months of time. RVPs stored at -80°C for lengths of times ranging from 2 days to 9 months were thawed and used to infect permissive cells (Figure 1). RVPs frozen for up to 9 months demonstrated equivalent infection to the same batch of RVPs frozen for only 2 days. RVPs also maintain infectivity at both 37°C and 4°C during the typical 1-2 h incubation with sera during neutralization and enhancement assays. RVPs from two different serotypes were tested for stability by pre-incubating the virions at the indicated temperatures for up to 9 hours, and then analyzing the RVPs for retention of infectivity (Figure 2). Results from these experiments demonstrate that RVPs retain stable infectivity under the experimental conditions used in most neutralization and enhancement assays.

Contact Us

Quality controlled Dengue Reporter Viruses are commercially available from Integral Molecular with defined strains of DENV serotypes 1, 2, 3, or 4 structural proteins, and with convenient luminescent reporters. For more information contact us at:

215.966.6061

info@integralmolecular.com

www.integralmolecular.com