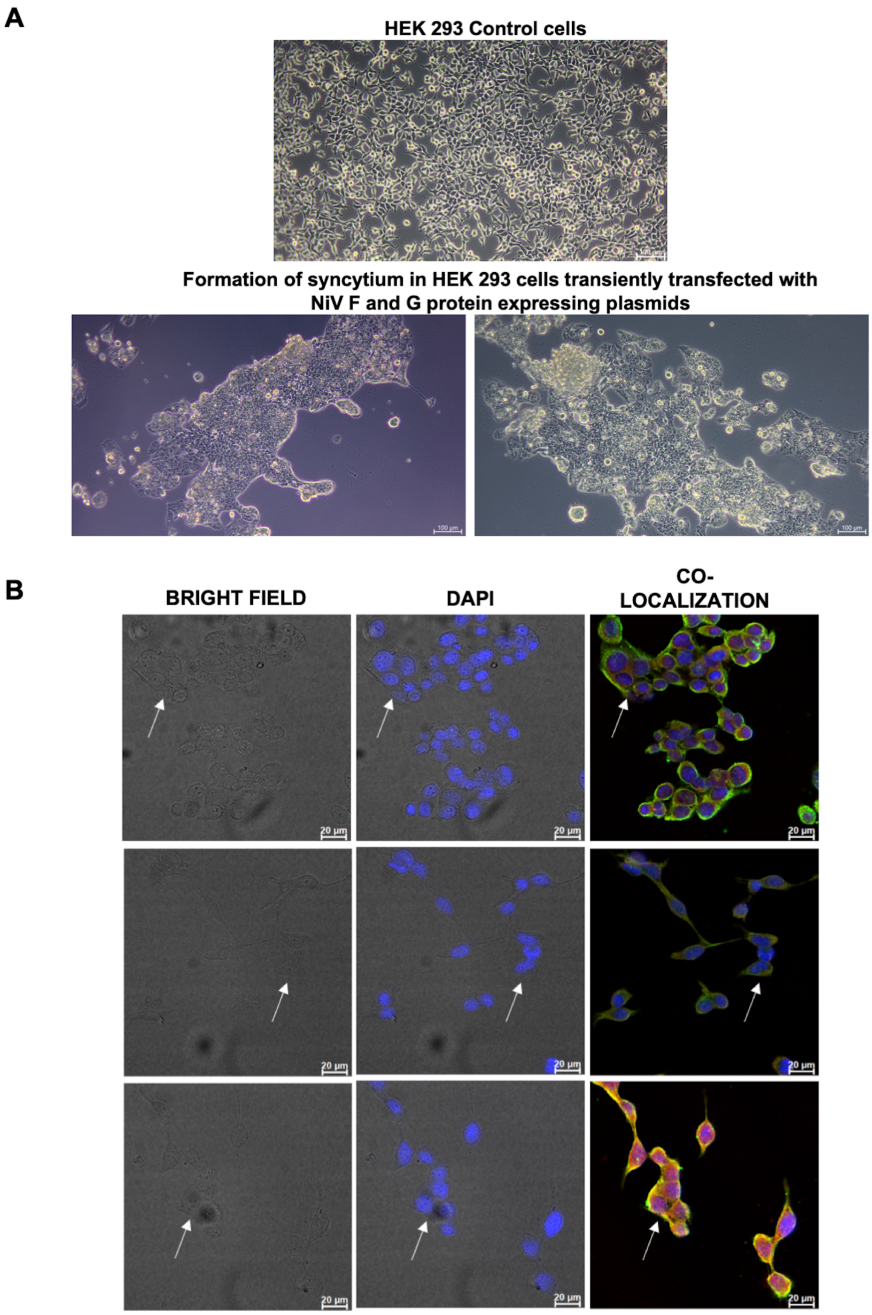
Reproducible generation of Nipah virus pseudovirions with uniform incorporation of F and G surface glycoproteins for high-throughput neutralization assays

Geetu Rose Varghese[1], Vivek Vijay[1], Sreeja Sreedevi[1], Santhik S Lupitha[1], Priya Prabhakaran[2], Sushama Aswathyraj[3], Anitha P. Moorkoth[4], Niyas K Pulloor[4] , Easwaran Sreekumar [1,5]*

[1]Molecular Bioassay Laboratory; [2]Department of Virus Applications; [3]Department of Molecular Diagnostics, Institute of Advanced Virology (IAV), Thonnakkal, Thiruvananthapuram 695317, Kerala, India.
[4]Department of Microbiology, Government Medical College, Kozhikode, Kerala, India.
[5]Molecular Virology Laboratory, Rajiv Gandhi Centre for Biotechnology (RGCB), Thiruvananthapuram 695014, Kerala, India

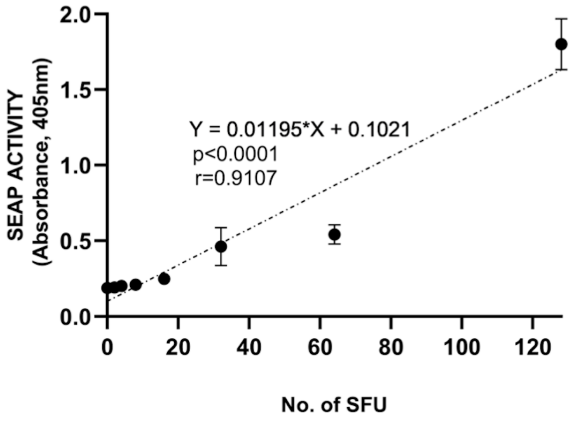
*Corresponding author: Easwaran Sreekumar
(E-mail: esreekumar@iav.res.in; esreekumar@rgcb.res.in)

Supplementary data



Supplementary Fig.1

Supplementary Fig.1. Immunofluorescence analysis of syncytia formation in HEK 293 cells upon transfection with NiV F and G proteins. A. HEK 293 cells transiently transfected with NiV F and G proteins depicting the formation of syncytial cells. Top panel: HEK293 control cells, Bottom panel: NiV F and G protein expressing plasmid transfected cells with large multinucleated syncytium**.** HEK293 cells expressing the plasmid- encoded NiV F and G proteins were fixed for immunofluorescence and stained with primary antibodies 12B2 and 48D3 respectively. Confocal microscopy revealed fused cells as indicated by the arrows.



Supplementary Fig. 2

Supplementary Fig.2. Titration of N-PV(FG)-5F6 pseudovirions by colorimetric Spot forming assay. Correlation analysis of Spot forming unit with SEAP assay. Serial dilutions of the virus inoculum with varying SFUs were used to infect BHK-21 cells and the SEAP activity was measured after 24h. The values are Mean±SD of two independent experiments done in triplicates (N=6).