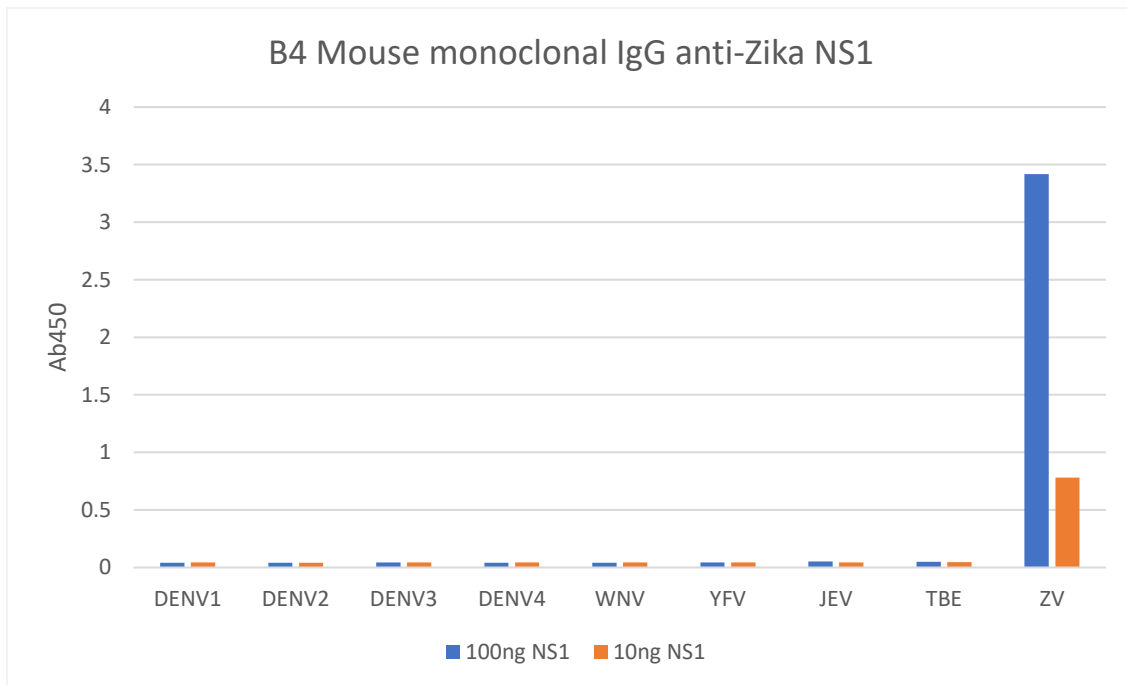


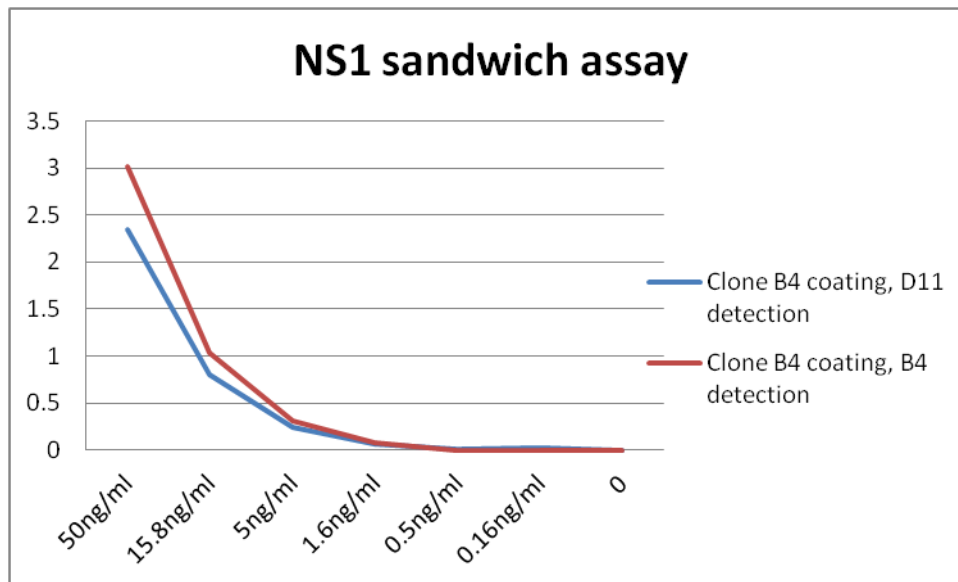
Direct ELISA data

An ELISA plate was coated with 10ng and 100ng of NS1 antigen per well, then blocked with 1% BSA. Primary antibody was used at a concentration of 1ug/ml, and the detection antibody used was Goat anti-mouse IgG:HRP (Bio-Rad, 1:2000). The substrate used was TMB (KPL).



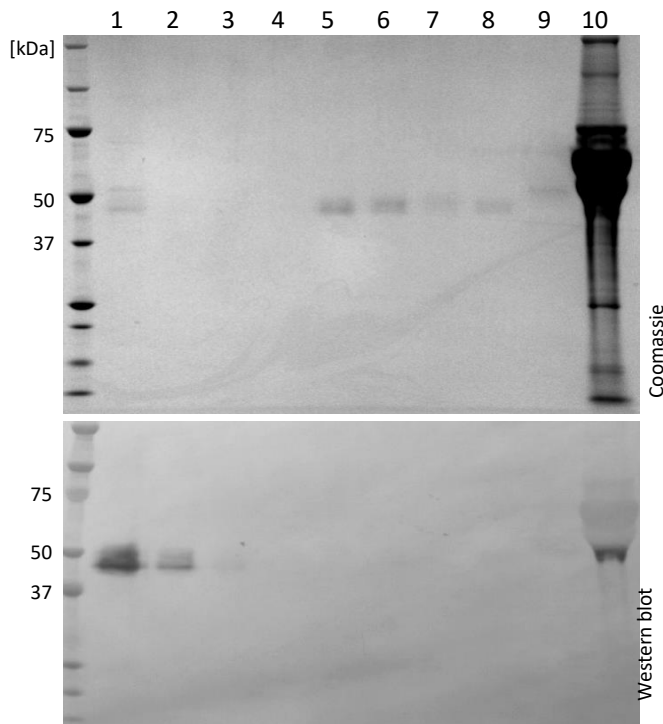
Sandwich Assay Data

An ELISA plate was coated with 500ng of coating antibody, then blocked with Casein, and samples were added at a range of concentrations. Detection antibody was biotinylated, and used at a concentration of 0.25ug/ml. Visualisation was with Streptavidin:HRP and the substrate used was TMB (KPL).



Western blot data

Variable amounts of antigen were loaded onto a NuPAGE 4-12% Bis-Tris gel (Life Technologies) and separated at 200V for 55 minutes. Western blot was performed using a Transblot Turbo system (Biorad) with pre-packed nitrocellulose membrane (Biorad). As blocking agent 5% non-fat milk powder in PBS-T was used. Antibodies were used at concentrations of 1ug/ml. Secondary antibody used was goat-anti-mouse-IgG-HRP (Bio-Rad). Western blots were developed using TMB Membrane (KPL).



- 1: Zika virus NS1 [100ng]
- 2: Zika virus NS1 [20ng]
- 3: Zika virus NS1 [4ng]
- 4: Zika virus NS1 [0.8ng]
- 5: Dengue virus serotype 1 [100ng]
- 6: Dengue virus serotype 2 [100ng]
- 7: Dengue virus serotype 3 [100ng]
- 8: Dengue virus serotype 4 [100ng]
- 9: West Nile virus NS1 NY99 [100ng]
- 10: supernatant of Zika virus infected Vero cells [5μl]

HH6.B10.A4.B4

Immunofluorescence data

Vero cells were seeded on coverslips infected with ZIKV (African or Asian strains) for 30h at MOI 0.5. Control coverslips consisted of uninfected Vero cells. After fixation with 4% PFA, samples were stained with Mouse anti-Zika virus NS1 (B4). Antibody was diluted 1:500 and Triton X-100 was used as detergent. Imaging was performed using a Leica SP5 confocal microscope (Virology Research Services). Immunofluorescence for both African and Asian strains was observed (African strain illustrated below).

