

Supplemental Figure legends

FIGURE S1. Platelet binding activity and anti-NS1 C-terminus titer of anti-NS1

Abs from mice immunized with 2, 3 or 5 doses of NS1. Polyclonal Abs were generated in mice after immunization with various doses of NS1 and IgG was purified on protein G columns. *A*, Platelets were incubated with Abs for 30 min, followed by FITC-conjugated anti-mouse IgG staining and analyzed by flow cytometry. The percentages of platelet binding were quantified. *B*, ELISA plate was coated with 0.5 mg/well of a mixture of NS1 C-terminal peptides (aa 271-280, 281-290, 291-309, 310-330 and 331-350) and various dilutions of Abs were added, followed by peroxidase-conjugated anti-mouse IgG. Data are presented as the mean \pm SD of triplicate cultures. * $p < 0.05$, ** $p < 0.01$.

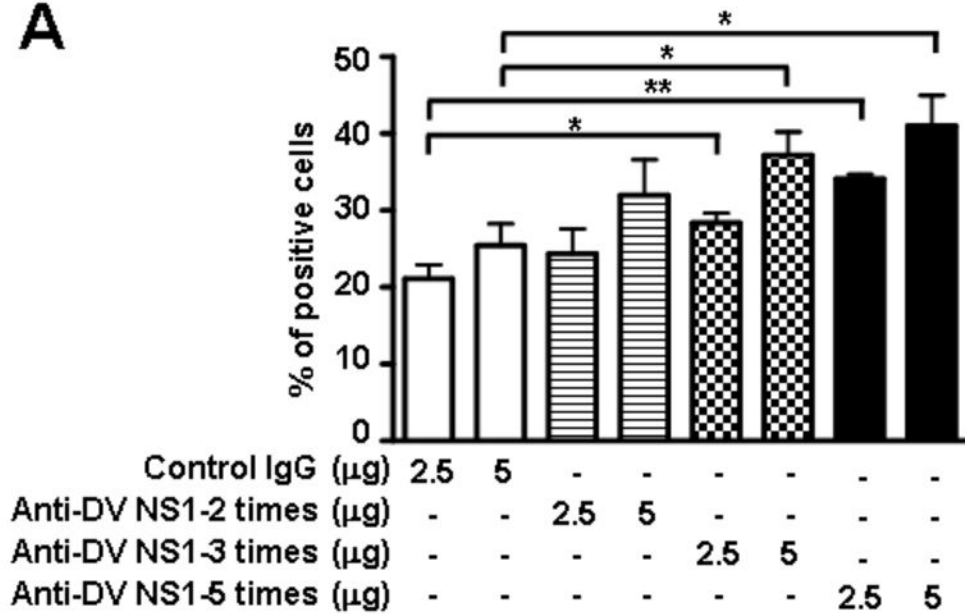
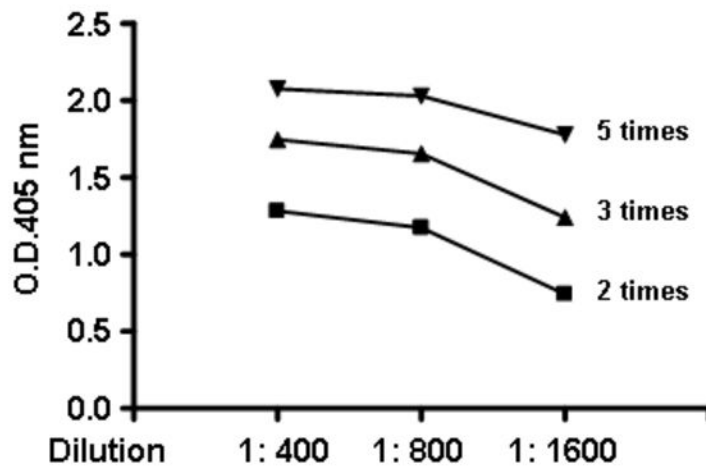
FIGURE S2. The bleeding times and platelet numbers in mice immunized with 2

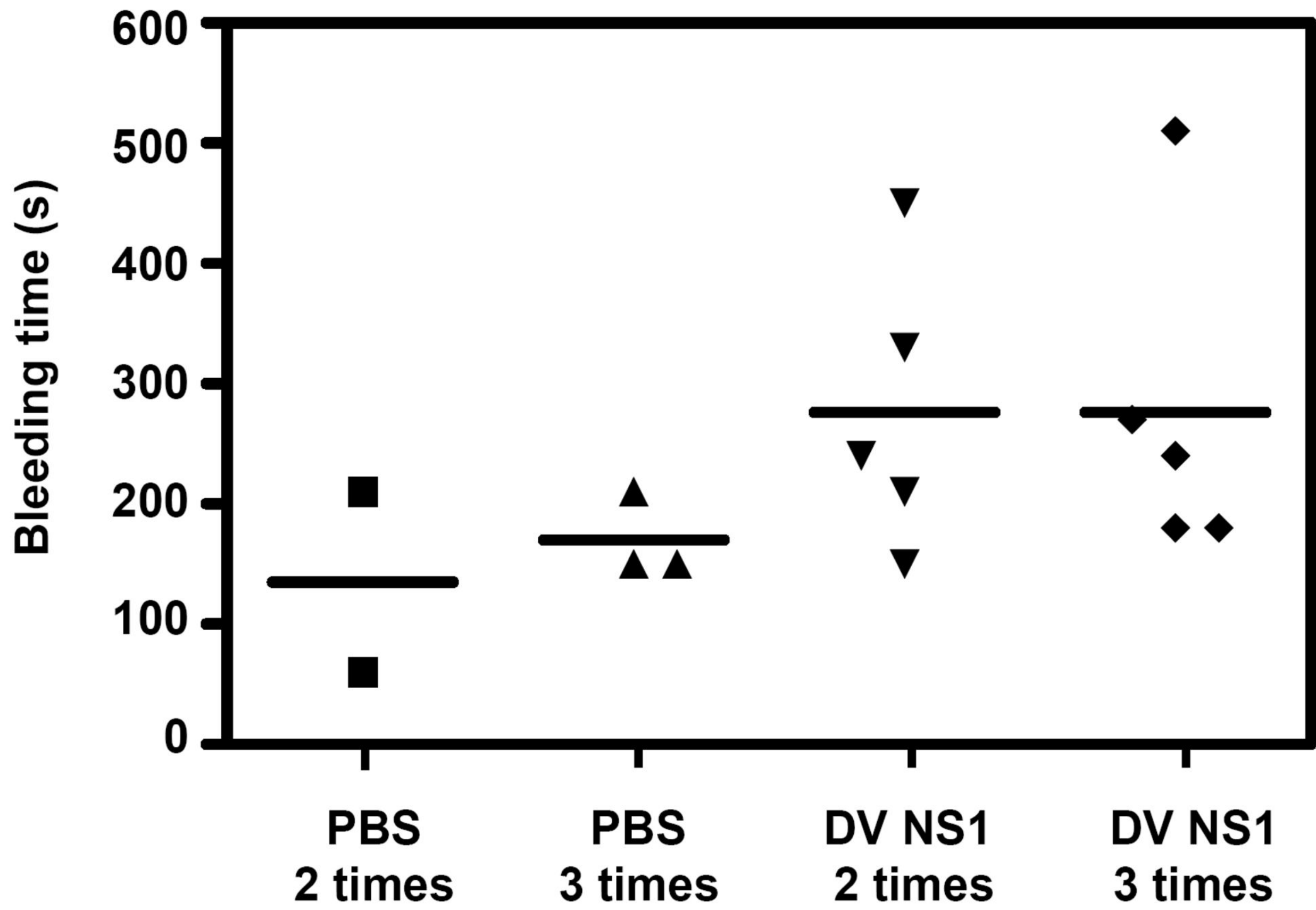
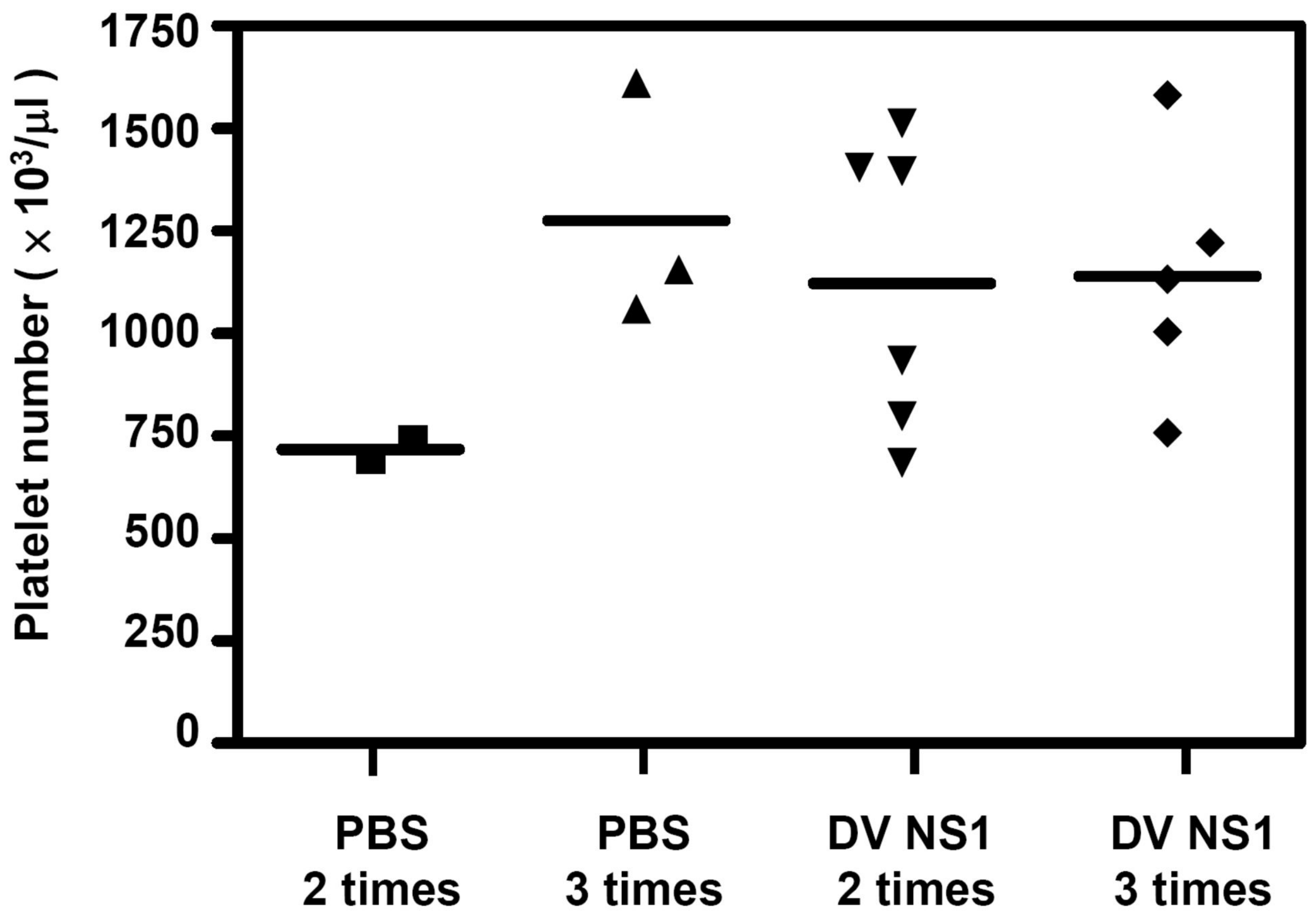
or 3 doses of NS1. C3H/HeN mice were i.p. immunized with PBS or DV NS1 in adjuvant for 2 or 3 times, and the bleeding times (*A*) and platelet numbers (*B*) were determined 3 days after the last injection.

FIGURE S3. Passively administered anti-NS1 Abs are diminished in mouse sera

and this decrease is correlated to the binding of Abs to platelets. Mice were i.v.

administered with 500 μ g of purified IgG obtained from mice after immunized with 2, 3 or 5 doses of DV NS1 ($n = 3$). A-C, After 1, 24, 48, and 72 h, blood samples were collected. The serum Ab titers were analyzed by ELISA. D, Freshly isolated mouse platelets after Ab administration for 24 h were washed and fixed with 1% formaldehyde in PBS, and then stained with FITC-conjugated anti-mouse IgG. Ab binding to platelets was detected and quantified by flow cytometry. $**p < 0.01$, $***p < 0.001$.

A**B**

A**B****Fig. S2**

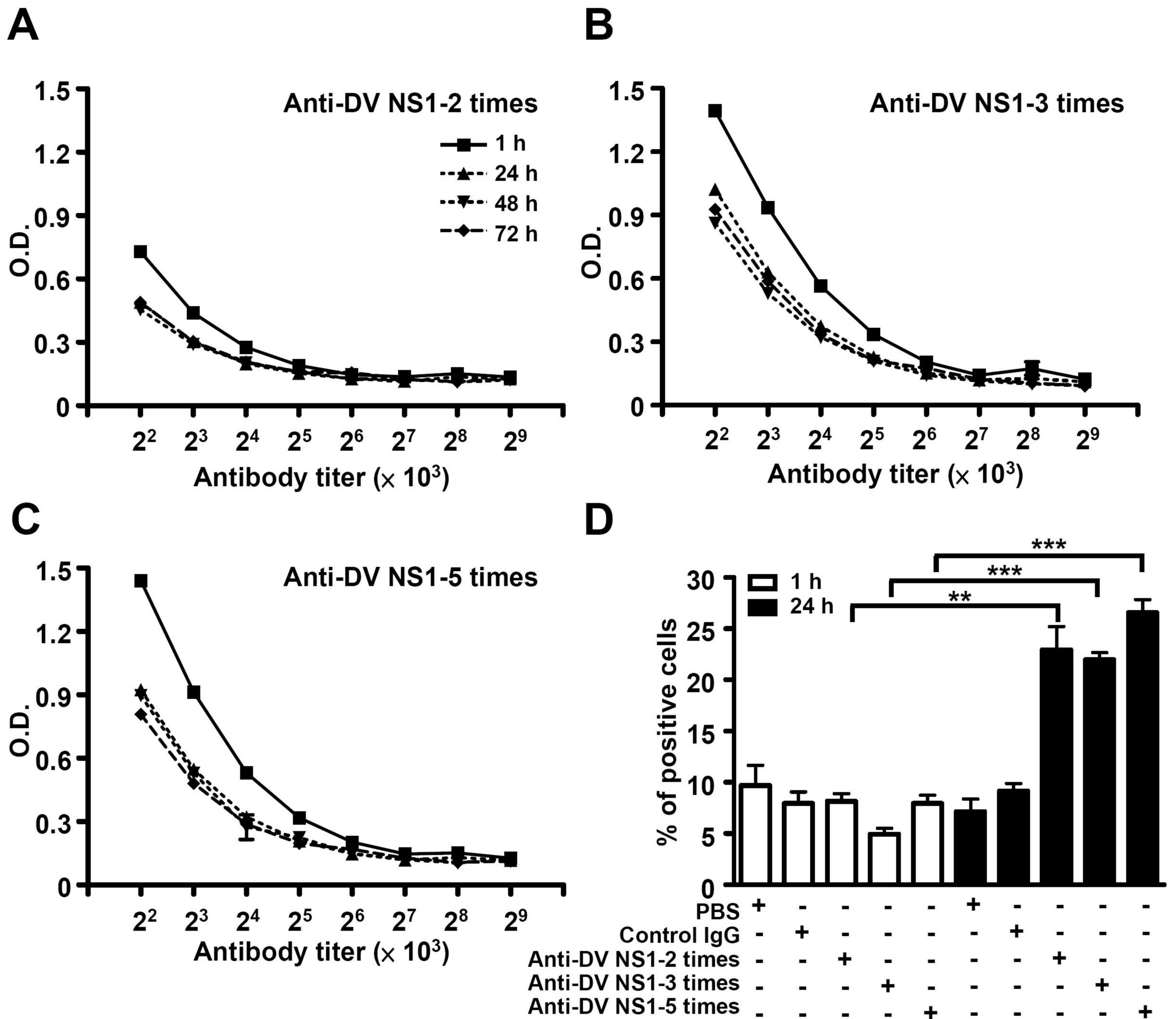


Fig. S3