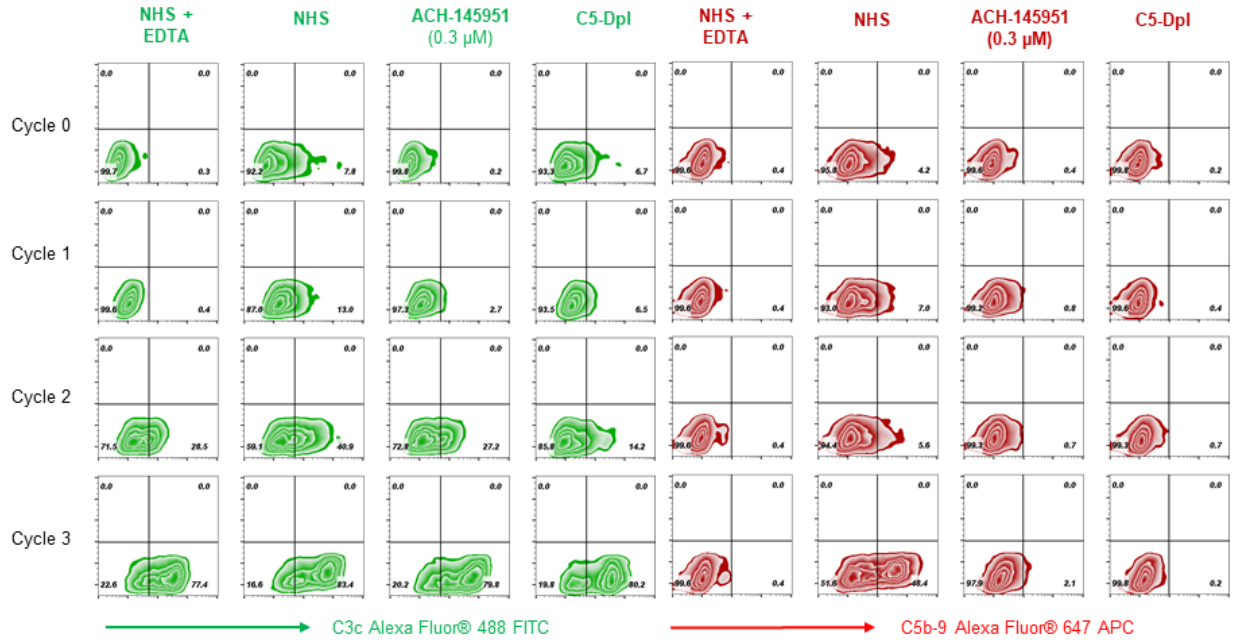
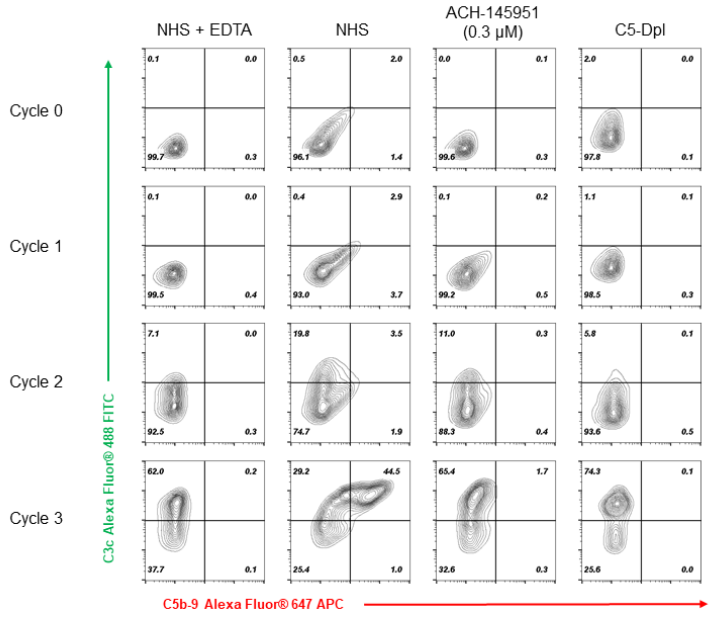
Supplemental appendix

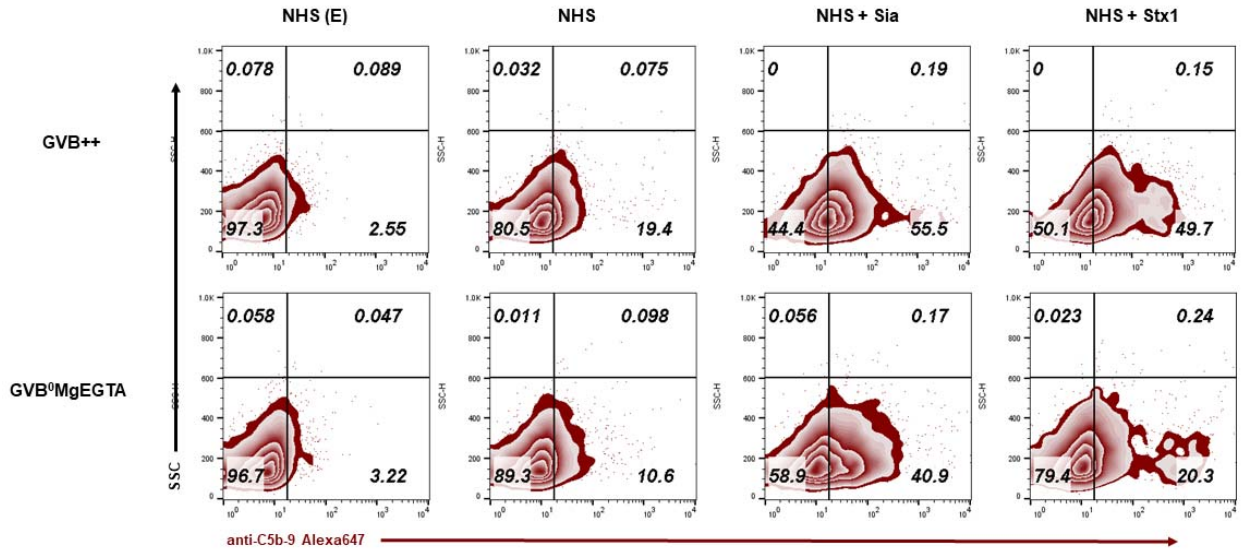
A.



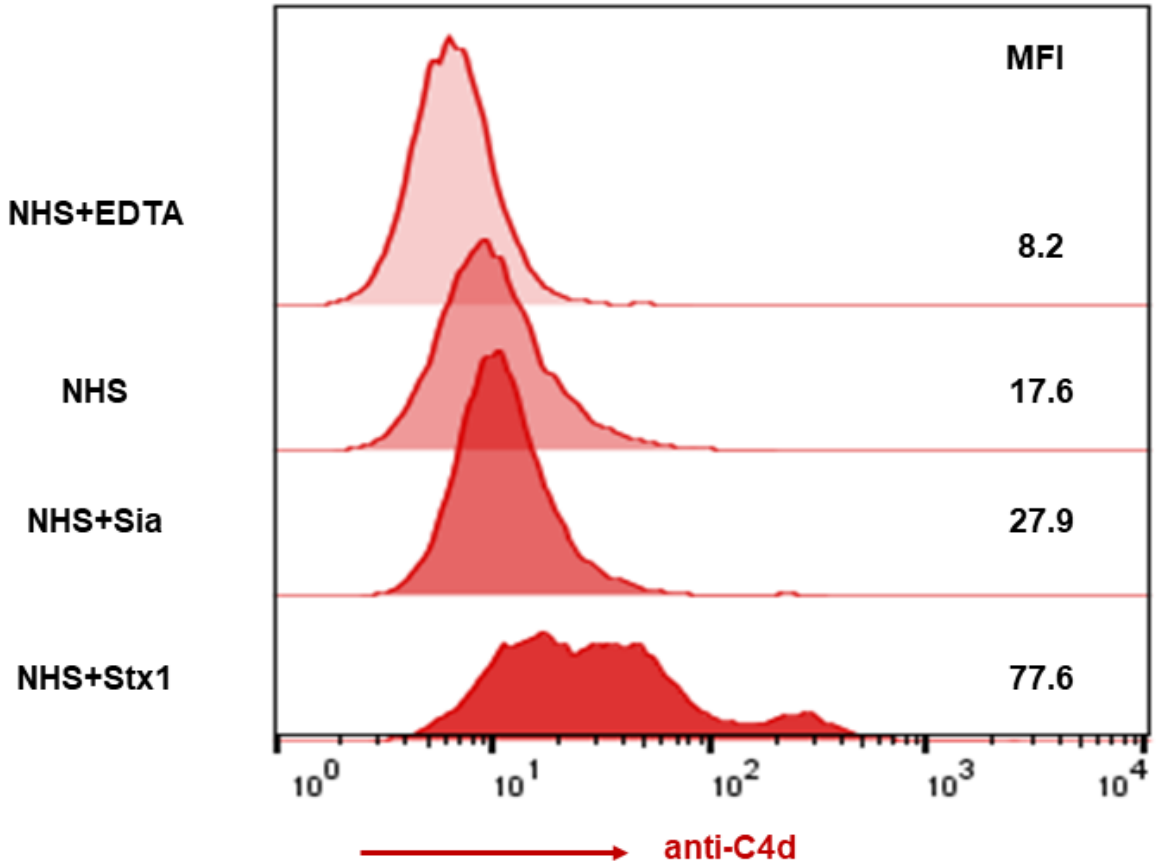
B.



Suppl. Fig. 1 C3b loading increases C5b9 deposition on TF1 *PIGA*null cells by flow cytometry. (**A**) Cells loaded with one to three cycles of C3b were incubated in 20% of NHS +EDTA, NHS, NHS + ACH-145951 and C5-depleted (C5-Dpl) serum. Zebra plots show $C3c^+$ (in green) on left graph and $C5b\text{-}9^+$ (in red) cells on the right graph. (**B**) Contour shows double positive for C3c (Y axis, Alexa 488) and C5b-9 (X axis, Alexa 647) (upper right). NHS +EDTA is negative control.



Suppl. Fig. 2 Comparison of C5b-9 deposition on TF1 *PIGA*null cells induced by Sialidase (Sia) and Shiga toxin 1 (Stx1) in GVB^{++} and $GVB^{0}EGTA$ buffers. FACS plot showing C5b-9 deposition following Sia and Stx1 treatment in GVB^{++} (top panel) and $GVB^{0}EGTA$ (bottom panel) buffers. The C5b-9 deposition (%) induced by Sia and Stx1 was similar (55.5% vs. 49.7%) in GVB^{++} buffer. However, the C5b-9 deposition caused by Stx1 (10 µg/ml) in $GVB^{0}EGTA$ buffer was reduced to 20.3%. NHS (E) normal serum + EDTA, NHS normal human serum.



Suppl. Fig. 3 Shiga toxin 1 (Stx1) induced C4d deposition on TF1 *PIGA*null cells in GVB^{++} by flow cytometry. TF1 *PIGA*null cells were treated with Stx1 or Sia in GVB^{++} buffer. C4d deposition was measured following Sia and Stx1 treatment. Overlaid histograms show that MFI of C4d was markedly enhanced with Stx1 compared to Sia. MFI (mean fluorescence intensity).