Effect of the protein corona on antibody-antigen binding in nanoparticle sandwich immunoassays

Supporting Information

Helena de Puig[1], Irene Bosch[2], Marc Carré Camps[3], and Kimberly Hamad-Schifferli[1,4]*

[1]Department of Mechanical Engineering, Massachusetts Institute of Technology, Cambridge, MA 02139

[2]Institute of Medical Engineering and Science, Massachusetts Institute of Technology, Cambridge, MA 02139

[3]Department of Chemical Engineering, Institut Quimic de Sarria, Universitat Ramon Llull, Via Augusta 390, 08017 Barcelona, Spain

[4]Department of Engineering, University of Massachusetts Boston, Boston, MA 02125

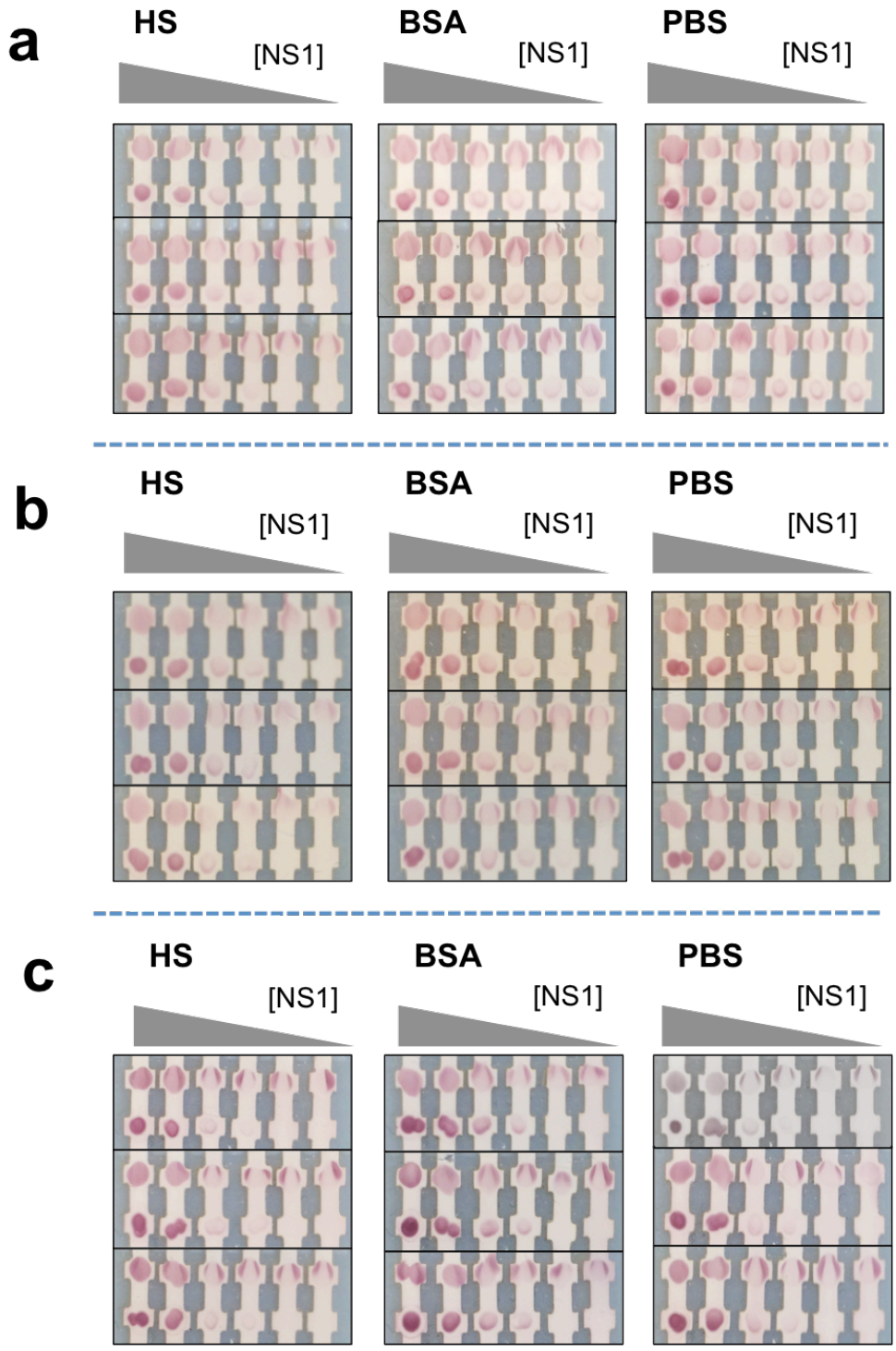


Figure S1. Triplicates of the dipstick immunoassay results for as a function of ZIKV NS1 concentration (L-R): 90.6 nM, 38.4 nM, 9.2 nM, 3.8 nM, 0.9 nM, 0 nM for a) NP-Abs in PBS, BSA, and HS, b) NP-Abs with preformed coronas in PBS, BSA, and HS; c) for test strips pre-treated with HS and then run in PBS, BSA, and HS.

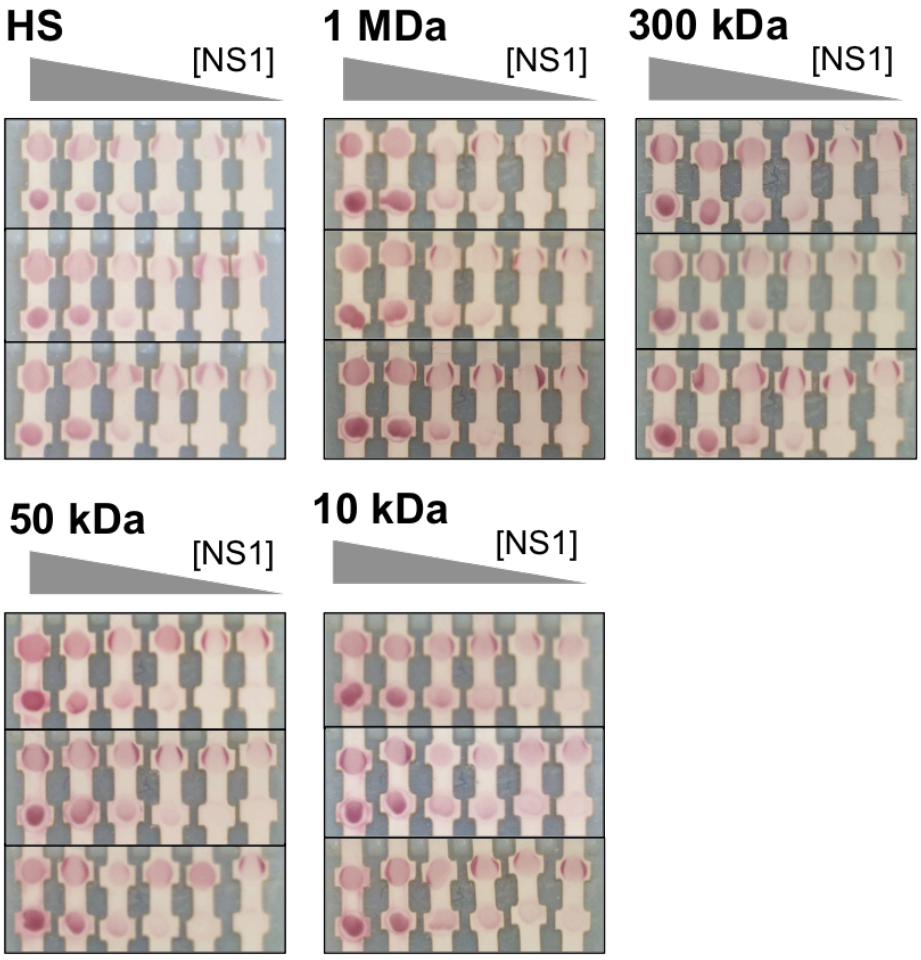


Figure S2. Triplicates of the dipstick immunoassay results as a function of ZIKV NS1 concentration (L-R): 90.6 nM, 38.4 nM, 9.2 nM, 3.8 nM, 0.9 nM, 0 nM for treatment of HS with Molecular weight cutoff (MWCO) filters of 1 MDa, 300 kDa, 50 kDa, 10 kDa and no filter (HS).

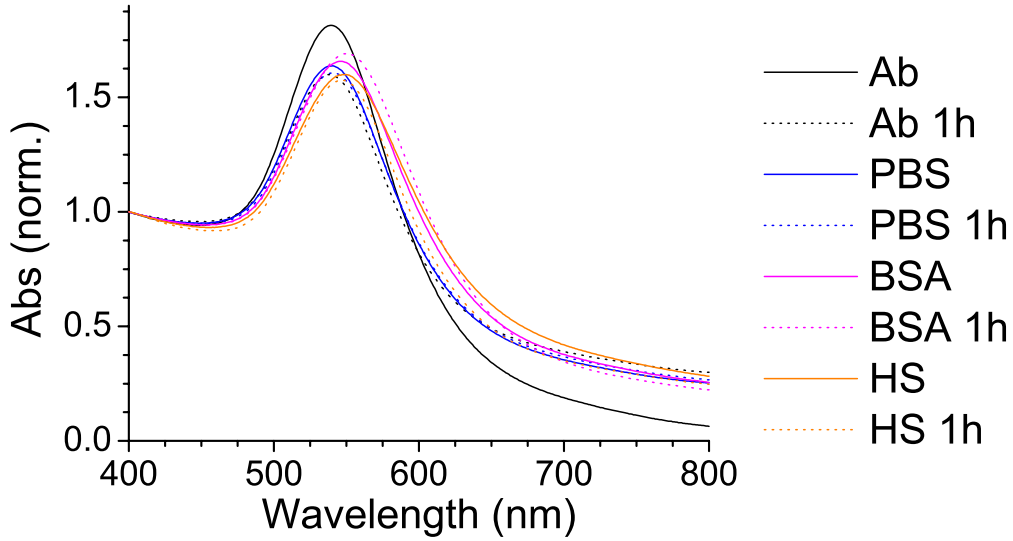


Figure S3. Optical absorption spectra of NP-Ab conjugates after incubation in the running buffers for 1h.

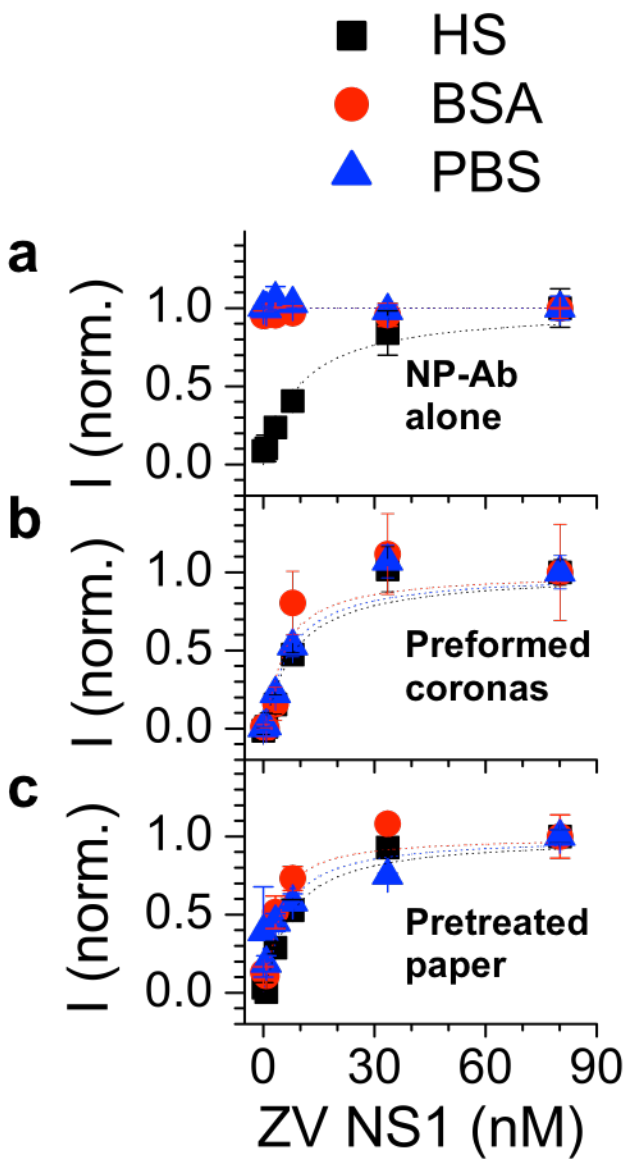


Figure S4. Non-covalently attached NP-Ab, strips and characterization. Test line intensity vs. NS1 concentration for NP-Abs in PBS (blue triangles), BSA (red circles), and HS (black squares), and fits to K_D^{eff} (lines). f) Test line intensity vs. NS1 concentration curves for NP-Abs with preformed protein coronas in PBS (blue triangles), BSA (red circles), and HS (black squares), and fits to K_D^{eff} (lines). g) Test line intensity vs. NS1 concentration curves for strips pretreated with HS and then run in in PBS (blue triangles), BSA (red circles), and HS (black squares), and fits to K_D^{eff} (lines).

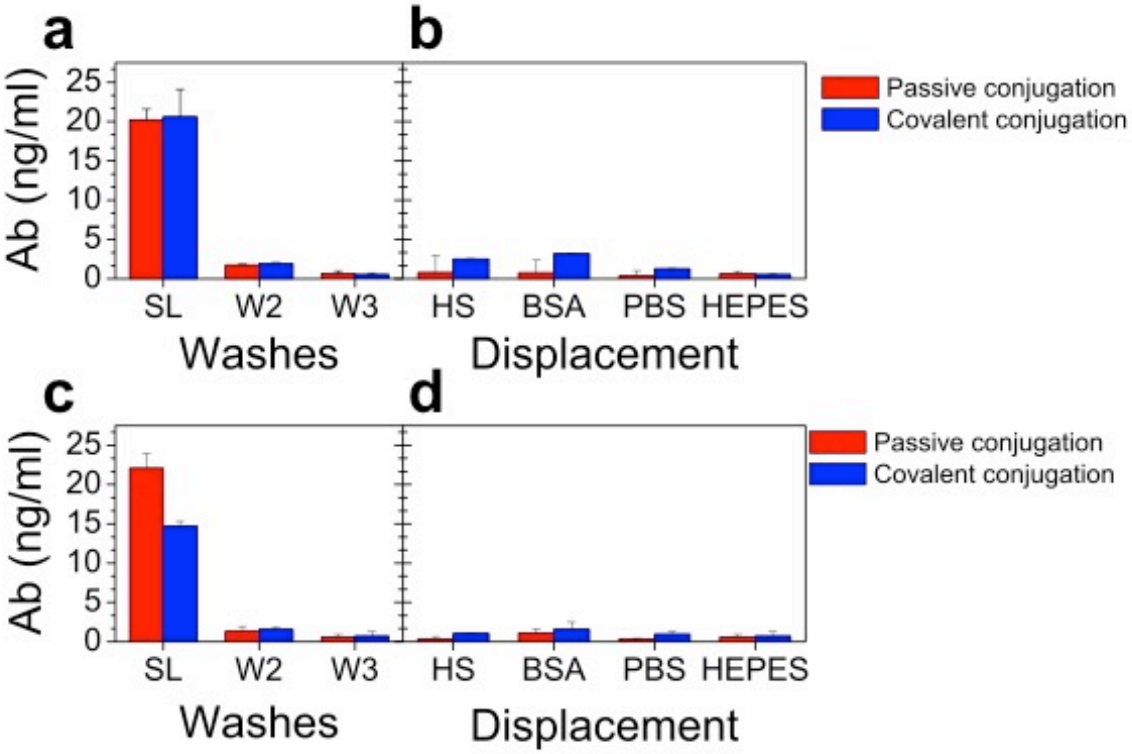
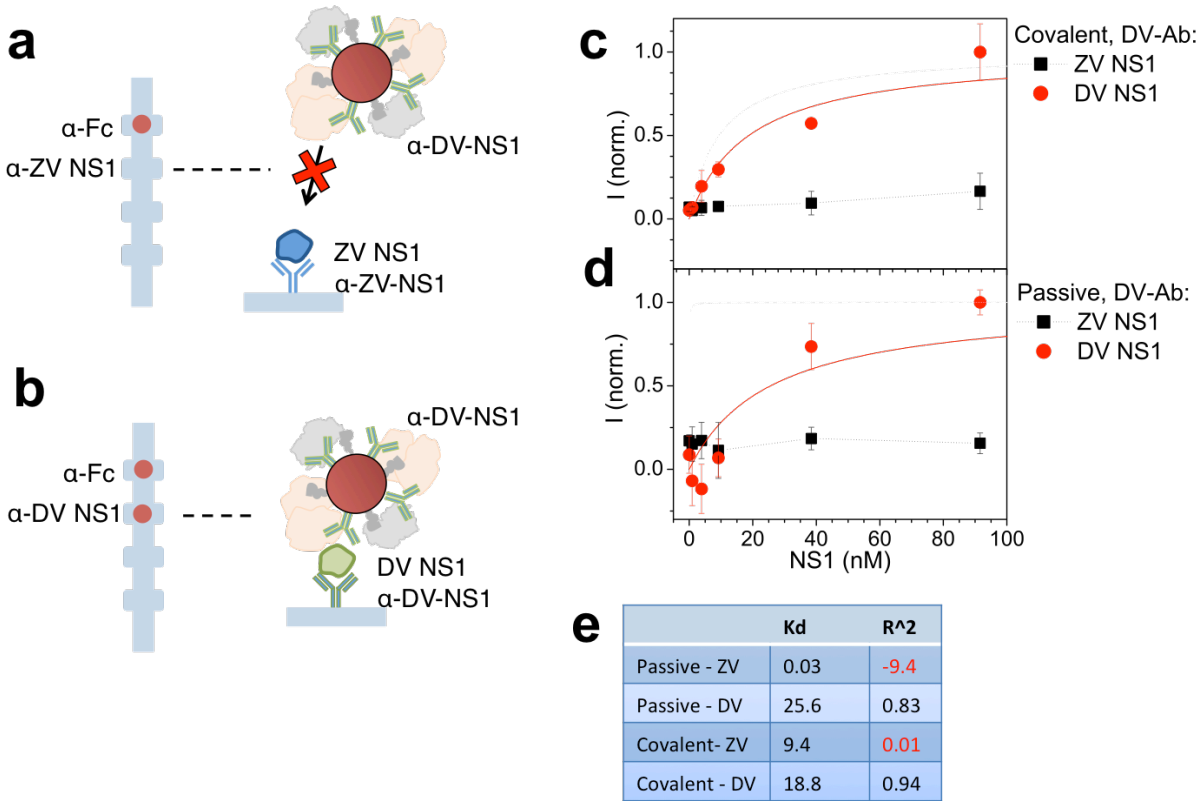


Figure S5: ELISA data for Ab displacement by HS for Non-covalent and covalent attachment. a) Measurement of Ab present in the washes after bioconjugation measured by ELISA. b) measurement of displacement of Abs when the purified NP-Ab conjugates are exposed to HS, BSA, PBS, and HEPES by ELISA. c) Measurement of Ab present in the washes after bioconjugation measured by fluorescence of fluorescently labeled Abs. d) measurement of displacement of Abs when the purified NP-Ab conjugates are exposed to HS, BSA, PBS, and HEPES by fluorescence of fluorescently labeled Abs.



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	Kd	R^2
Passive - ZV	0.03	-9.4
Passive - DV	25.6	0.83
Covalent- ZV	9.4	0.01
Covalent - DV	18.8	0.94

Figure S6. Control experiment of dipstick assays with dengue antibodies and dengue NS1 probing involvement of NS1 in corona formation. A) dipstick assay with immobilized anti-Zika antibodies and NPs conjugated to anti-Dengue antibodies run with Zika NS1, b) dipstick assay with immobilized anti-Dengue antibodies and NPs conjugated to anti-Dengue antibodies run with Dengue NS1, c) Test line intensity of the NP-anti-DV run with Zika NS1 (black squares) and NP-anti-DV run with Dengue NS1 (red circles) for covalently attached anti-DV antibodies, d) Test line intensity of the NP-anti-Dengue run with Zika NS1 (black squares) and NP-anti-Dengue run with Dengue NS1 (red circles) for non-covalently attached anti-Dengue antibodies, e) K_Ds from fits of the binding curves.