

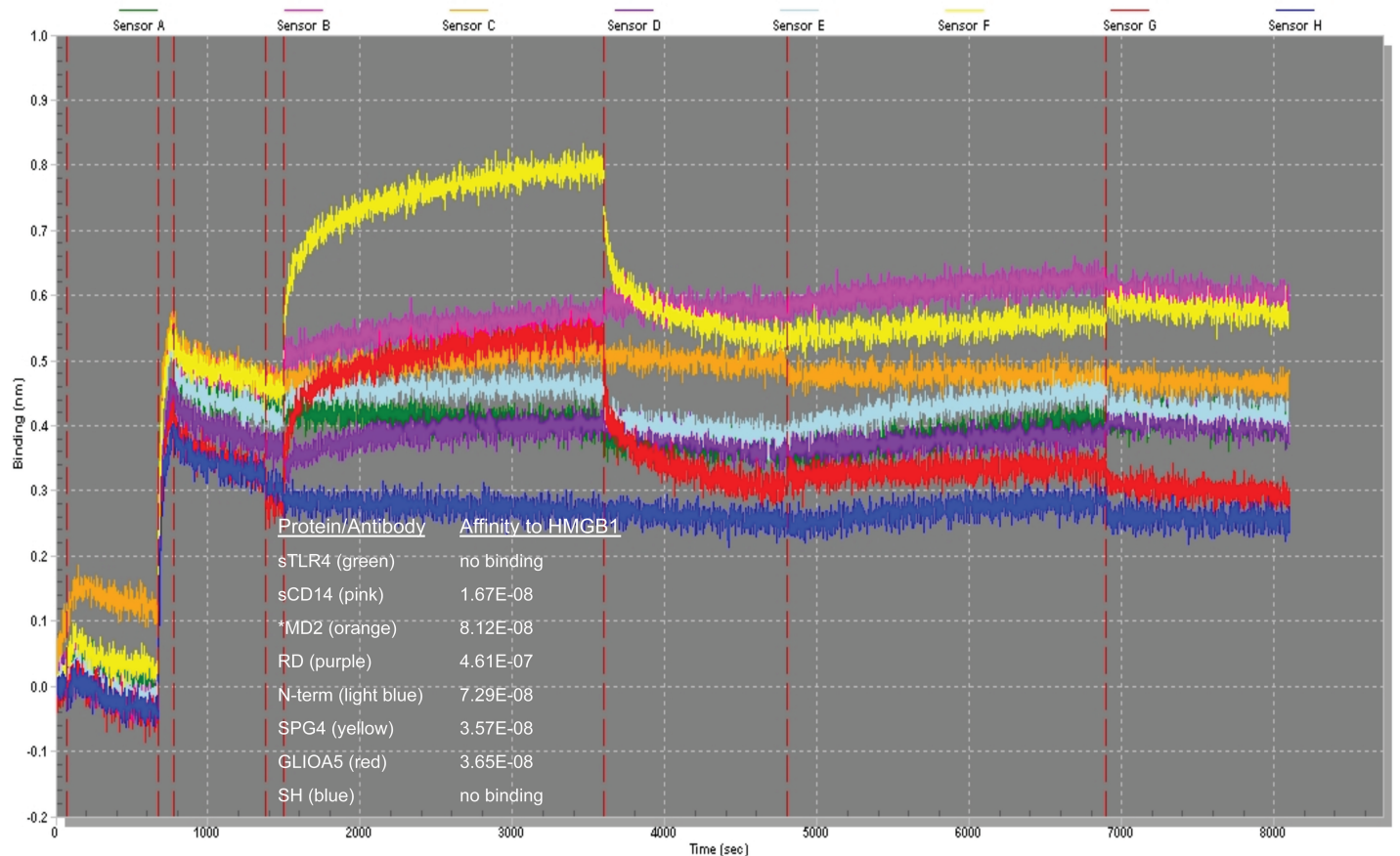
Supplemental Data

Signaling of High Mobility Group Box 1 (HMGB1) through Toll-like Receptor 4 in Macrophages Requires CD14

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**Supplementary Figure S1.** HMGB1 directly binds sCD14. Amine biosensors were loaded with solutions of 25 µg/mL of HMGB1 and then introduced into solutions containing nanomolar concentrations of sCD14 (pink), TLR4 (green) and MD2 (orange). Spastic paraplegia 4 (SPG4; yellow) was used as a positive binding control. RD (purple), 5H (dark blue), N-term (light blue) and GLIOA5 (red) are all generated anti-HMGB1 antibodies that were also tested for HMGB1 binding affinity. Dissociation was assessed by washing biosensors in PBS. The association and dissociation curves are depicted for a representative assay that has been controlled for the sample with kinetics buffer alone.