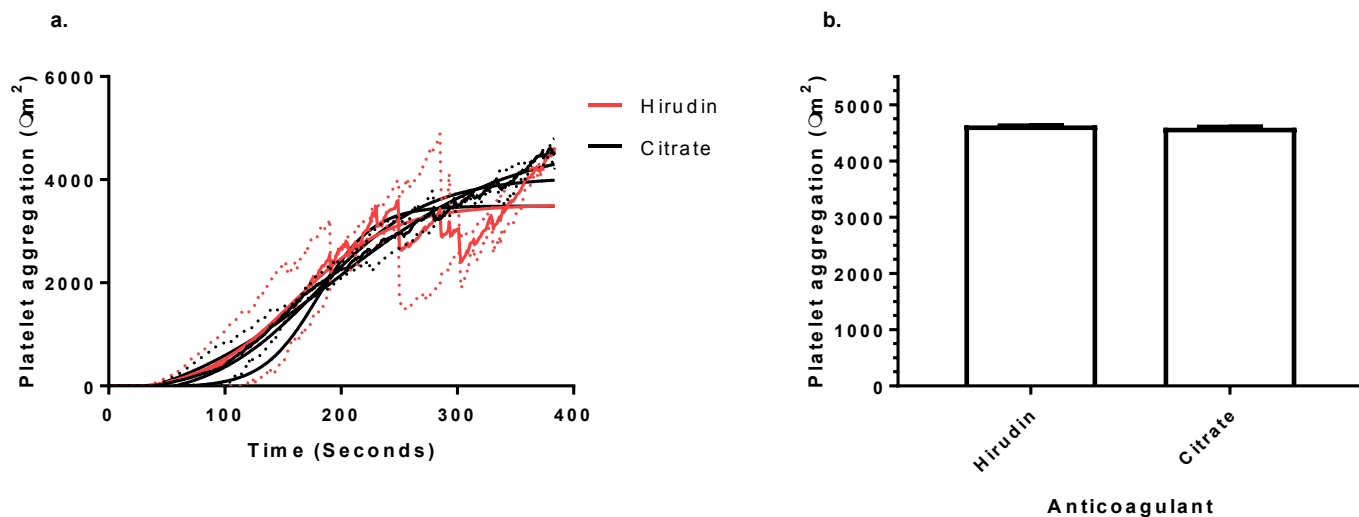


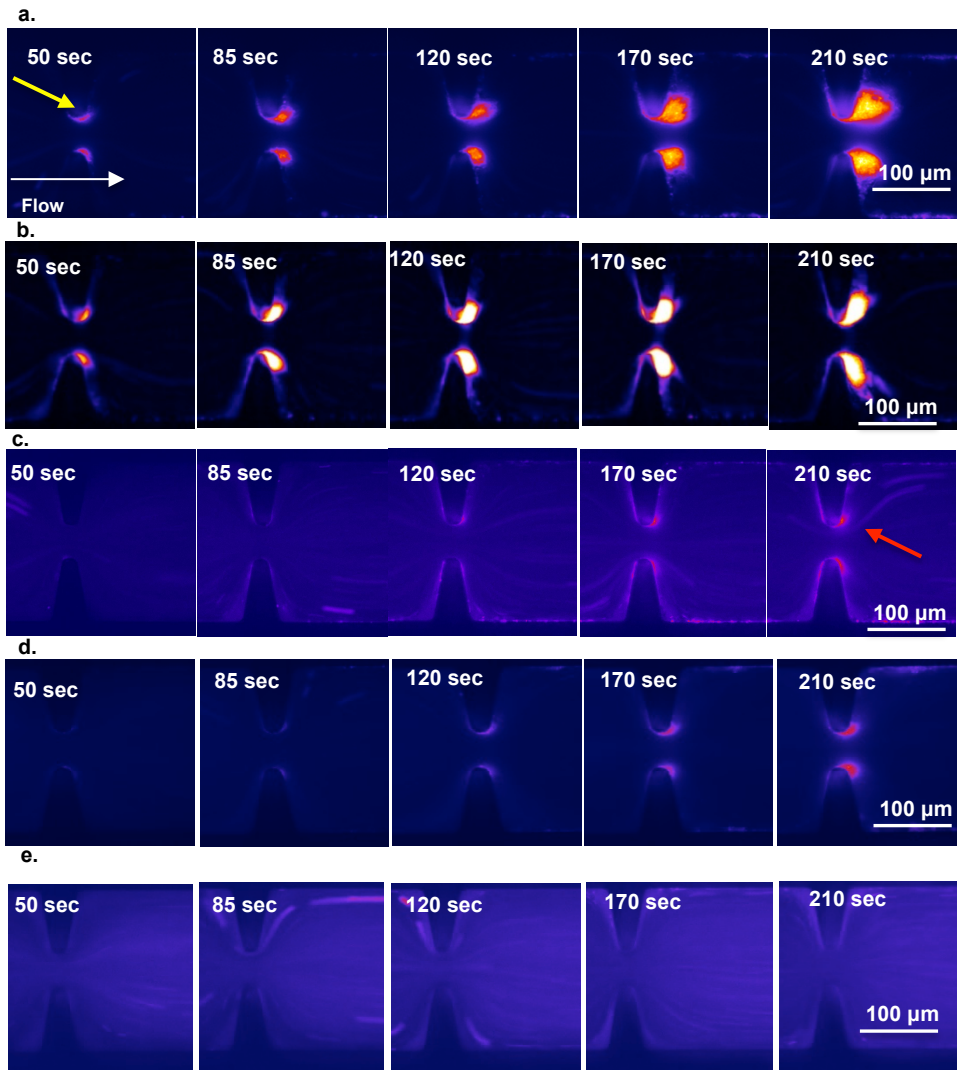
Supplementary Figure 1



Supplementary Figure 1. Anticoagulant comparison

a. Representative aggregate growth vs time graph showing platelet aggregation after collection in 3.2% citrate (black) or hirudin (80 U/mL) (red). Whole blood treated with MRS2179 (100mM), 2MeSAMP (10mM), Indomethacin (10 mM), and apyrase (1 U/mL) for 10 min @ 37°C prior to device perfusion (n = 2). Nonlinear curve fitting and 95% CI shown. **b.** Maximal aggregate growth over 3.5 min of device perfusion (n=2 donors).

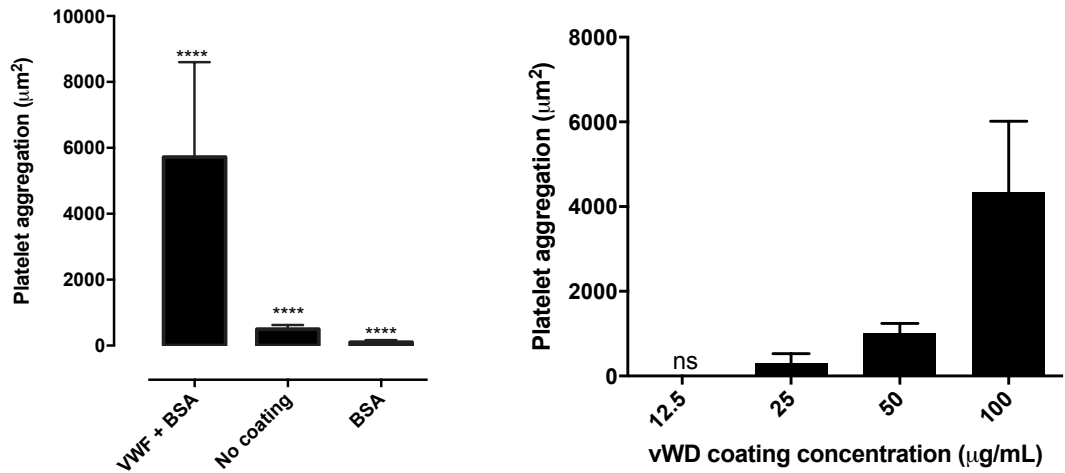
Supplementary Figure 2



Supplementary Figure 2. Device description and characterization

a. Representative timelapse images of platelet aggregation dynamics at the axisymmetric device microcontraction showing the overall development of the platelet aggregate in citrated whole blood. Yellow arrow denotes the point of aggregate initiation at the downstream margin of the 15μm long apex. Note that semistable aggregates project into the flow deceleration zone where aggregate surface shear rates approach $0.s^{-1}$. Also note the axisymmetric aggregation. **b.** Representative timelapse images of platelet aggregation dynamics at the axisymmetric device microcontraction showing the overall development of the platelet aggregate in amplification loop blocked (ALB) blood. **c.** Representative timelapse images of platelet aggregation in a type 1 VWD patient. Red arrow denotes the decreased maximal aggregate area seen in VWD samples. **d.** Representative timelapse images of platelet aggregation in a type 2 VWD patient. **e.** Representative timelapse images of platelet aggregation in a type 3 VWD patient.

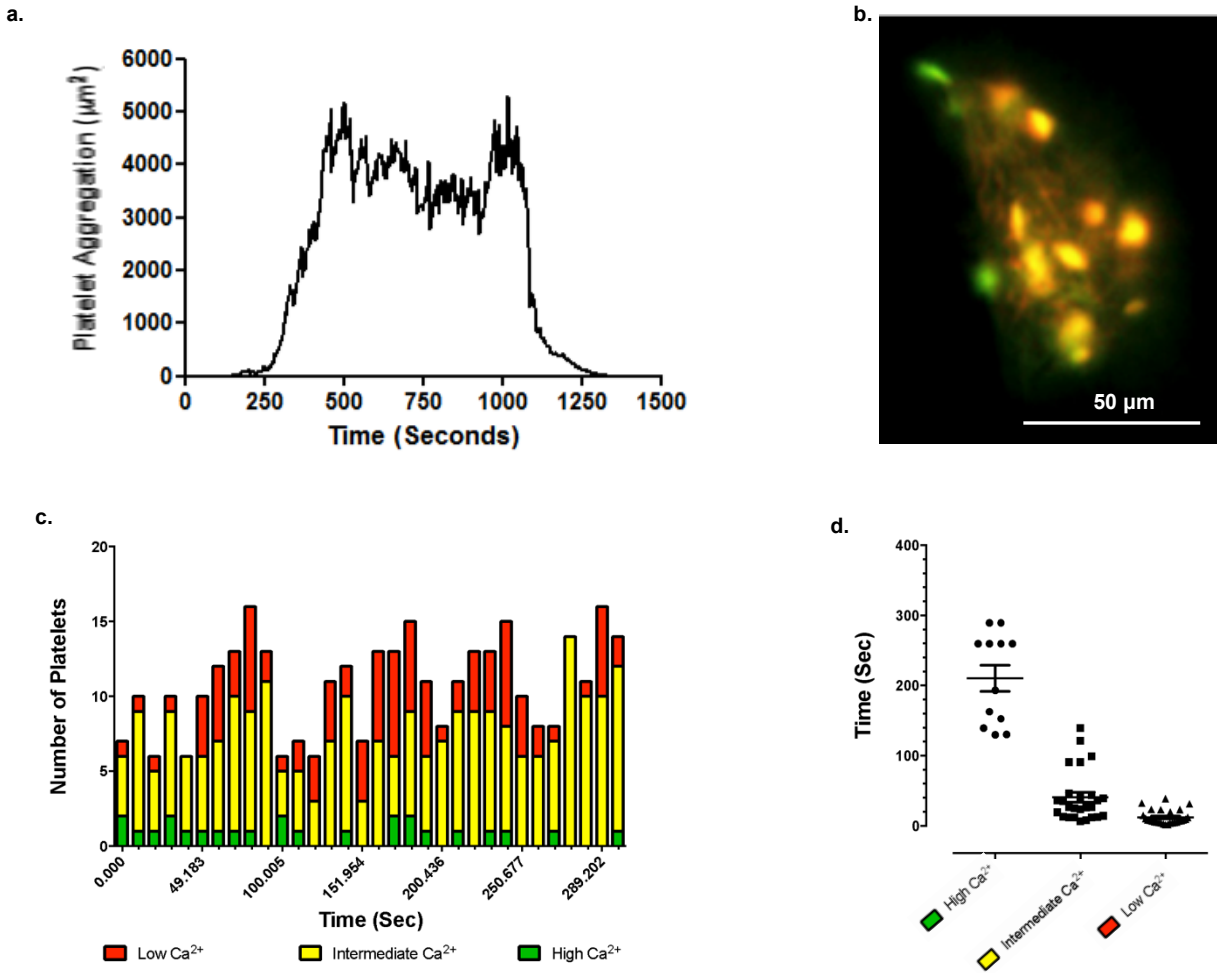
Supplementary Figure 3



Supplementary Figure 3. Device Performance as a Function of Coating Method

a. Maximal platelet aggregation as a function of coating method. VWF + BSA - device treated with 100 $\mu\text{g/mL}$ VWF followed by 10 $\mu\text{g/mL}$ BSA (as per Materials & methods); No coating – whole blood perfused through naïve device without any prior protein coating; BSA – device coated with 10 $\mu\text{g/mL}$ BSA alone prior to blood perfusion (n= 5 experiments). **b.** Device performance as a function of VWF coating concentration 12.5 – 100 $\mu\text{g/mL}$. Note that maximal aggregation maps to VWF concentration (n=3 experiments).

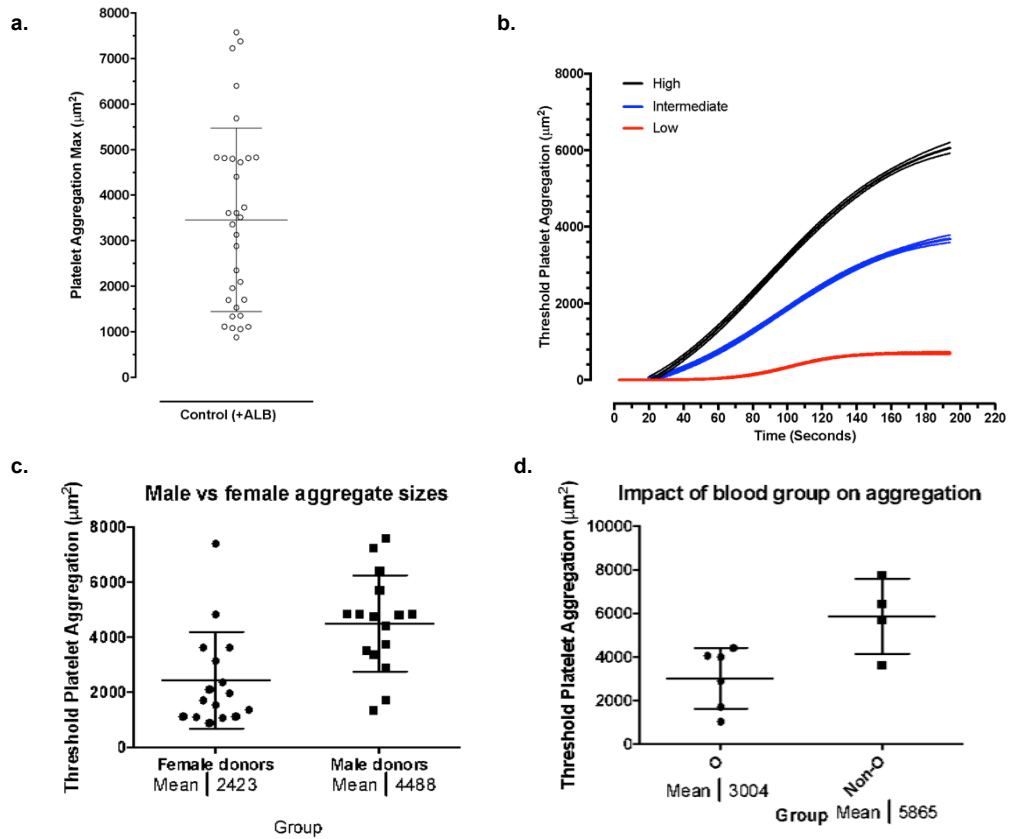
Supplementary Figure 4



Supplementary Figure 4. Platelet activation Status & Aggregate Dynamics

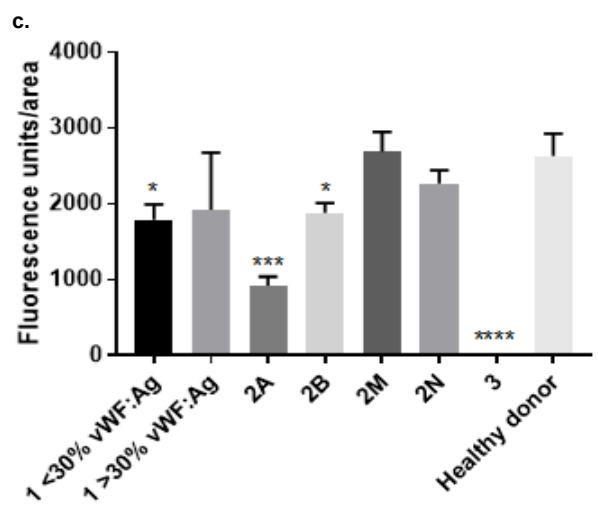
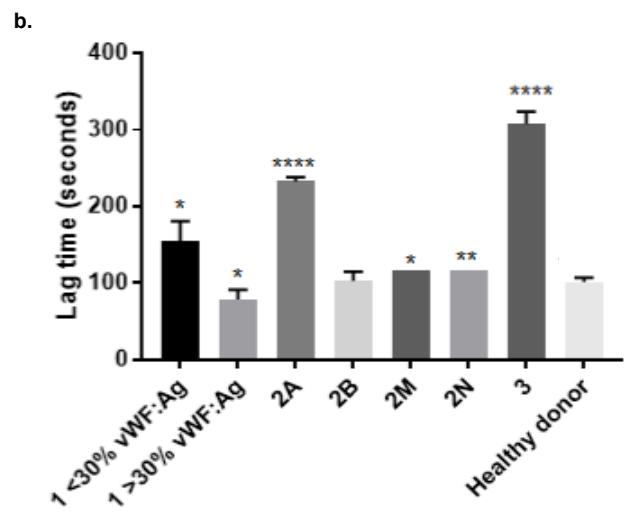
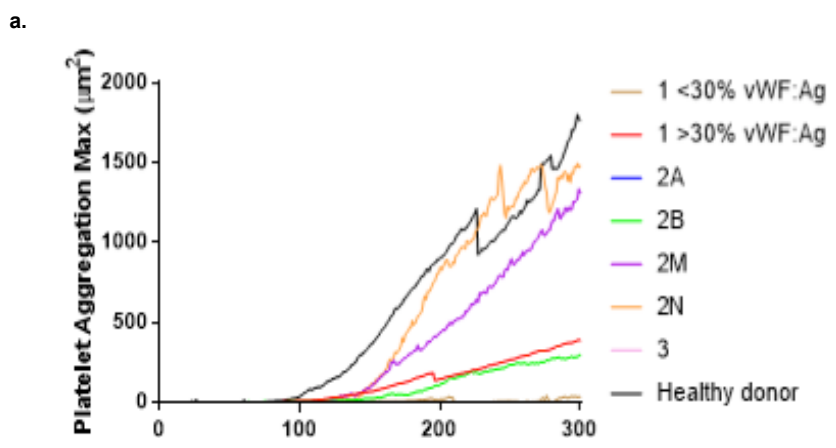
a. Representative aggregation trace showing chase experiments in which ALB treated DiOC6 labelled whole blood was chased with unlabeled whole blood samples at $t=1125$ s ($n=3$). Note that complete platelet turnover is achieved post chase demonstrating that the aggregate is in a state of dynamic equilibrium. **b.** Representative image showing the overall $[Ca^{2+}]_c$ distribution within a developing platelet aggregate downstream of the device microcontraction geometry (representative of $n = 3$ experiments). Note that High $[Ca^{2+}]_c$ are found primarily at the VWF derivitised PDMS substrate. **c.** Frequency distribution of platelet $[Ca^{2+}]_c$ as a function of time in developing platelet aggregates in the device ($n = 3$ experiments). **d.** Dot plot showing the relationship between time in aggregate and platelet $[Ca^{2+}]_c$. Note that platelets showing significant levels of Ca^{2+} flux demonstrate the greatest aggregate retention time. Also note that aggregates are primarily composed of platelets in a intermediate to low activation state.

Supplementary Figure 5



Supplementary Figure 5. Device performance

a. Platelet aggregation v time graphs showing device sensitivity to varying Hct levels. Citrated whole blood treated with MRS2179 (100mM), 2MeSAMP (10mM), Indomethacin (10 mM), and apyrase (1 U/mL) for 10 min @ 37°C prior to device perfusion (n = 3). Nonlinear curve fitting and 95% CI shown. **b.** Maximal platelet aggregation \pm SD following 3.5 min of device perfusion (n = 3 donors). **c.** Platelet aggregation v time graphs showing device sensitivity to varying platelet count ($\times 10^9$). Citrated whole blood treated with MRS2179 (100mM), 2MeSAMP (10mM), Indomethacin (10 mM), and apyrase (1 U/mL) for 10 min @ 37°C prior to device perfusion (n = 3). Nonlinear curve fitting and 95% CI shown. **d.** Maximal platelet aggregation \pm SD following 3.5 min of device perfusion (n = 3 donors).



Supplementary Figure 6. Device performance across VWD subtypes

a. Maximal platelet aggregation ± SD following 3.5 min of device perfusion. Citrated whole blood treated with MRS2179 (100mM), 2MeSAMP (10mM), Indomethacin (10 mM), and apyrase (1 U/mL) for 10 min @ 37°C prior to device perfusion. **b.** Lag time until initial platelet adhesion, as measured by time until first fluorescence detection vs VWD subclassification. **c.** Maximal fluorescence per unit area of aggregate vs VWD subclassification, as measured by determining maximal aggregate size and level of fluorescence at that point.