Supplementary Information

Cationic PAMAM dendrimers disrupt key platelet functions

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Supplemental Table 1. Physicochemical Characterization of PAMAM Dendrimers

Probe	FITC moieties per dendrimer	Zeta potential (mV)* labeled dendrimer	Zeta potential (mV) unlabeled dendrimer#
G7NH2-FITC	8.9 ± 1.0	23.5 ± 1.3	64.8 ± 3.2#
G6.5COOH-FITC	2.6 ± 0.5	-34.7 ± 6.0	-42 ± 1.2#
G7OH-FITC	1.2 ± 0.7	16.2 ± 0.2	27.7 ± 1.1#

All values are represented as Mean ± S.D. (n=3)

*Measured at pH 7.4 (not buffered), 25°C

#Previously published in reference 27

Supplemental Video 1. Time-lapse video (DIC) depicting increased platelet adherence to fibrinogen coated microfluidic wells under flow after G7-NH_2 dendrimer treatment. As in Figure 6, platelets were left untreated or stimulated with thrombin (0.5 U/mL) or G7-NH_2 dendrimer (100 μg/mL) and flowed at 200 s^{-1} for 6 minutes over Ibidi 0.4 VI plates pre-coated with 0.5 mg/mL fibrinogen. Images were captured in real-time on an Olympus wide-field fluorescent microscope (IX-81 inverted microscope system) using an ORCA-ER monochrome CCD camera (capture rate 1Hz). The arrow in the upper left hand corner indicates the direction of the flow. Each video is one experiment, representative of twelve independent experiments.

Supplemental Video 2. Time-lapse video (epifluorescence) depicting increased platelet adherence to fibrinogen-coated microfluidic wells under flow after G7-NH_2 dendrimer treatment. As in Figure 6, platelets were left untreated or stimulated with thrombin (0.5 U/mL) or G7-NH_2 dendrimer (100 μg/mL) and flowed at 200 s^{-1} for 6 minutes over Ibidi 0.4 VI plates pre-coated with 0.5 mg/mL fibrinogen. Platelets were fluorescently labeled before treatment. Images were captured in real-time on an Olympus wide-field fluorescent microscope (IX-81 inverted microscope system) using an ORCA-ER monochrome CCD camera (frame capture rate 1 Hz). The arrow in the upper left hand corner indicates the direction of the flow. Each video is one experiment, representative of nine independent experiments.

Supplemental Video 3. Time-lapse video (epifluorescence) depicting $\alpha_{IIb}\beta_3$ dependence of thrombin-treated platelet adherence to fibrinogen-coated microfluidic wells. As in Figure 6, platelets were stimulated with thrombin (0.5 U/mL) for five minutes and flowed at 200 s^{-1} for 6 minutes over Ibidi 0.4 VI plates pre-coated with 0.5 mg/mL fibrinogen. Platelets were pre-treated for 30 minutes with abciximab (0.136 μM), Fab fragment against $\alpha_{IIb}\beta_3$, and fluorescently labeled before treatment with agonist. Abciximab-treated platelets had reduced adhesion to fibrinogen, independent of agonist treatment. Images were captured in real-time on an Olympus wide-field fluorescent microscope (IX-81 inverted microscope system) using an ORCA-ER monochrome CCD

camera (frame capture rate 1 Hz). The arrow in the upper left hand corner indicates the direction of flow. Each video is one experiment, representative of six independent experiments.

Supplemental Video 4. Time-lapse video (epifluorescence) depicting $\alpha_{IIb}\beta_3$ dependence of G7-NH_2-treated platelet adhesion to fibrinogen-coated microfluidic wells. As in Figure 6, platelets were stimulated with G7-NH_2 dendrimer (100 μg/mL) and flowed at 200 s^{-1} for 6 minutes over Ibidi 0.4 VI plates pre-coated with 0.5 mg/mL fibrinogen. Platelets were pre-treated for one hour with abciximab (0.136 μM), Fab fragment against $\alpha_{IIb}\beta_3$, and fluorescently labeled before treatment with agonist. Abciximab-treated platelets had reduced adhesion to fibrinogen independent of agonist treatment. Images were captured in real-time on an Olympus wide-field fluorescent microscope (IX-81 inverted microscope system) using an ORCA-ER monochrome CCD camera (frame capture rate 1 Hz). The arrow in the upper left hand corner indicates the direction of flow. Each video is one experiment, representative of six independent experiments.