

# **Model Particulate Drug Carriers Modulate Leukocyte Adhesion in Human Blood**

## **Flows**

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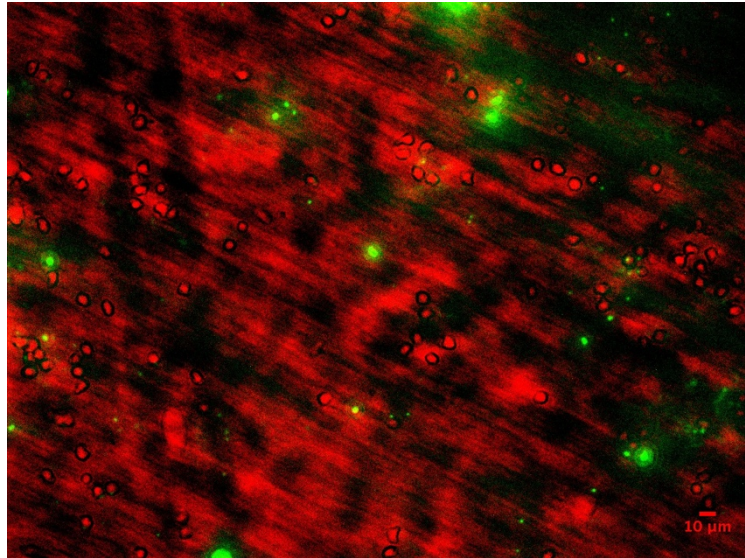
<sup>+</sup>Department of Chemical Engineering, University of Michigan, Ann Arbor, MI 48109

## **Supporting Information**

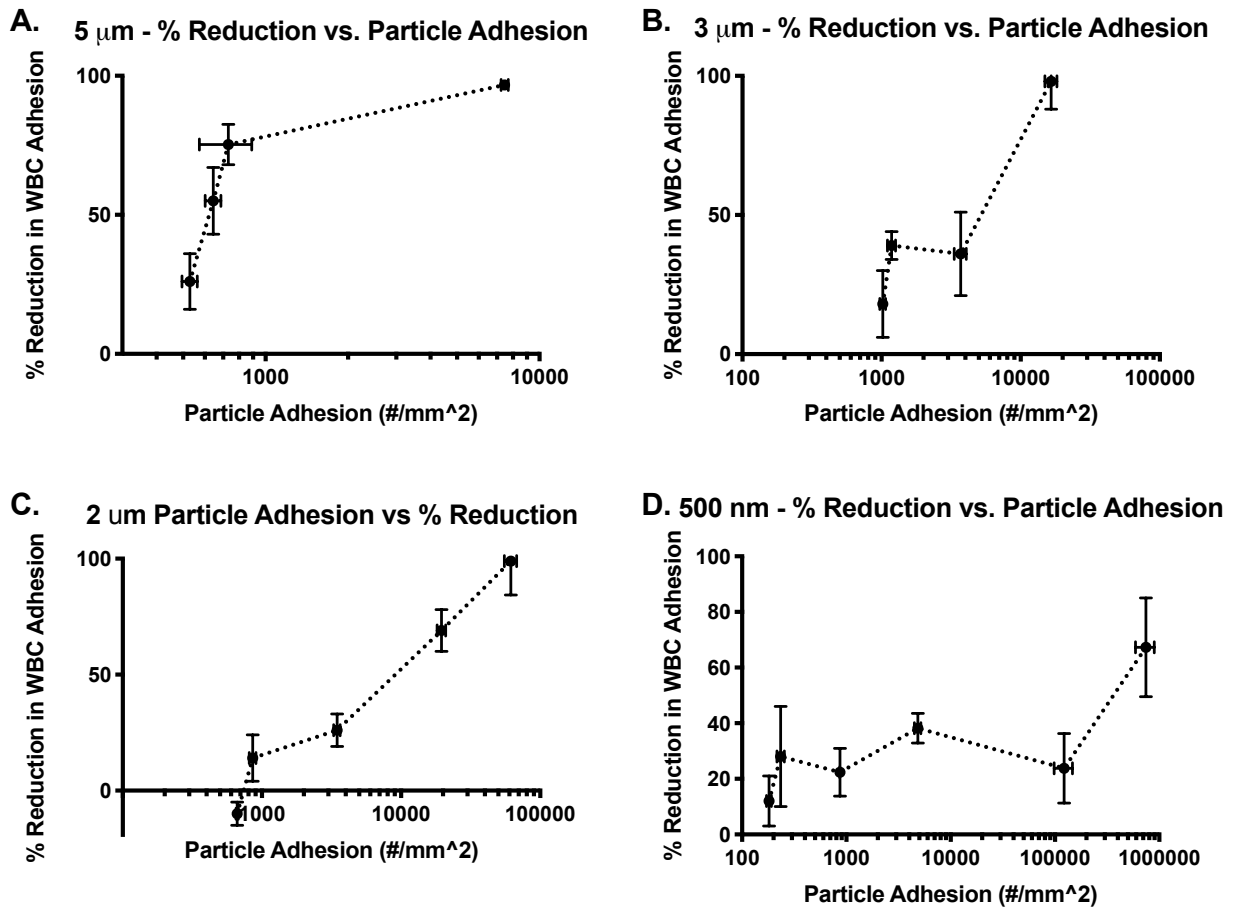
**Pages in Supporting Information: 9**

**Figures in Supporting Information: 8**

## Supporting Information

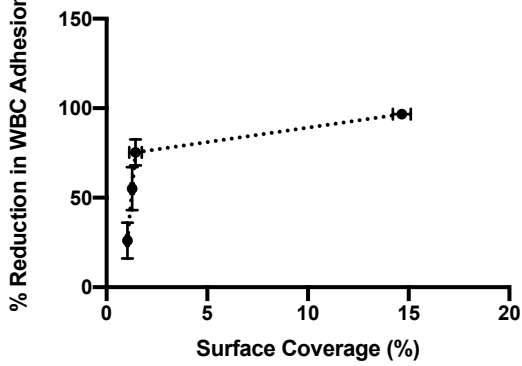


**Supplemental Figure 1. Microscope image of 500 nm IgG-conjugated fluorescent particles co-localizing with neutrophils on an inflamed endothelium. Red = CD45-PE, green = FITC particles.**

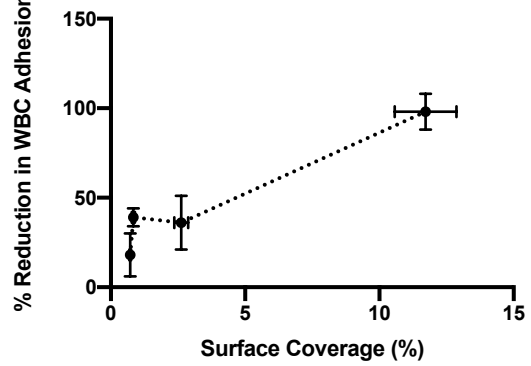


**Supplemental Figure 2. Particle Adhesion versus Percent Reduction in Leukocyte Adhesion.** (a) Particle Adhesion vs. % Reduction in Leukocyte Adhesion for 5  $\mu\text{m}$  particles. (b) Particle Adhesion vs. % Reduction in Leukocyte Adhesion for 3  $\mu\text{m}$  particles. (c) Particle Adhesion vs. % Reduction in Leukocyte Adhesion for 2  $\mu\text{m}$  particles. (d) Particle Adhesion vs. % Reduction in Leukocyte Adhesion for 200 nm particles. Each data point represents at least 3 independent donors. Error bars are standard error.

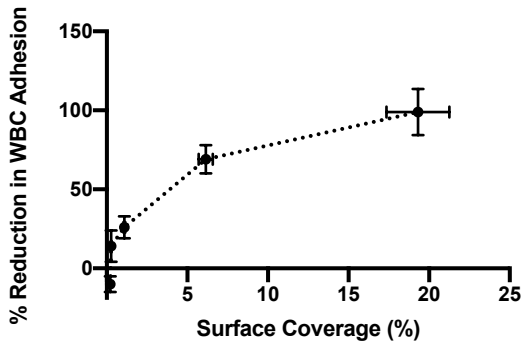
**A. 5  $\mu\text{m}$  - % Reduction vs. Surface Coverage**



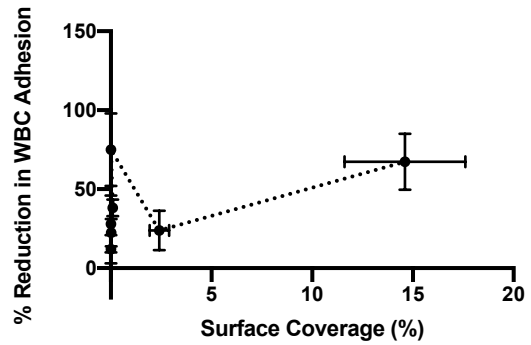
**B. 3  $\mu\text{m}$  - % Reduction vs. Surface Coverage**



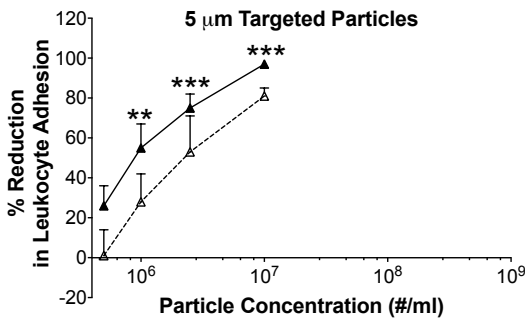
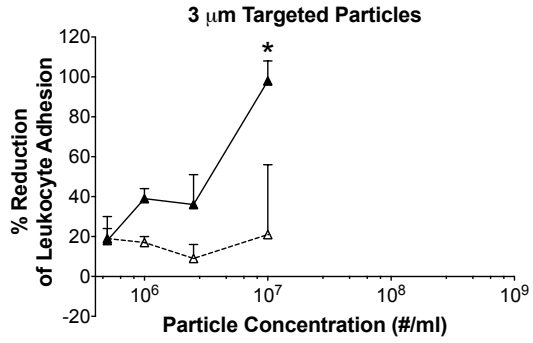
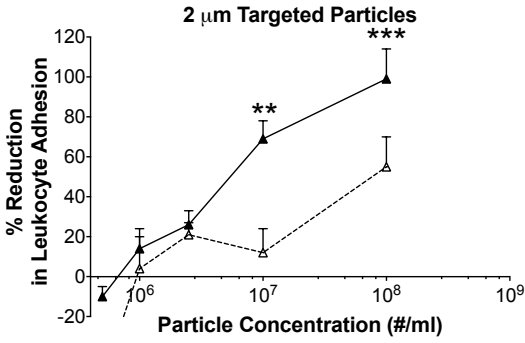
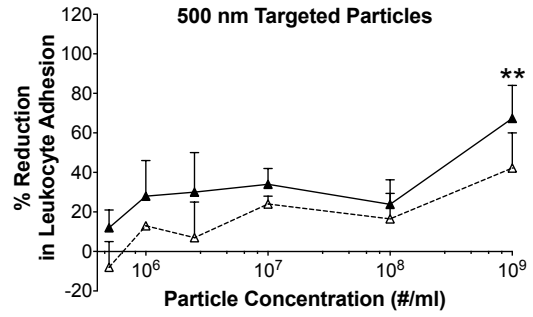
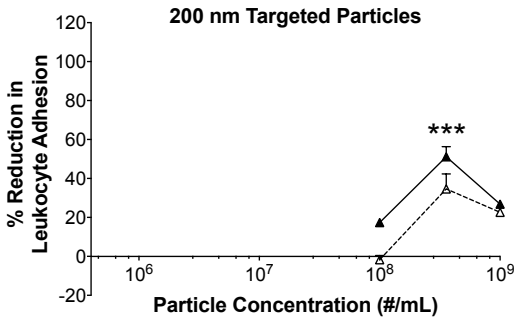
**C. 2  $\mu\text{m}$  - % Reduction vs. Surface Coverage**



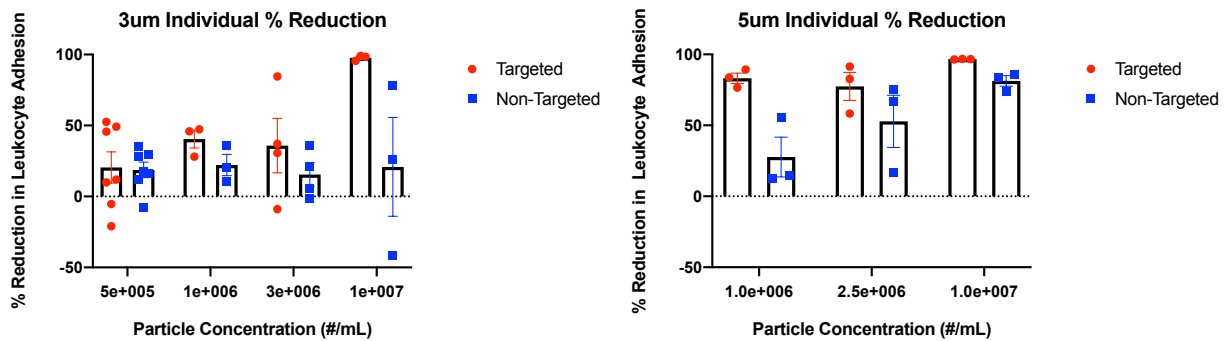
**D. 500 nm - % Reduction vs. Surface Coverage**



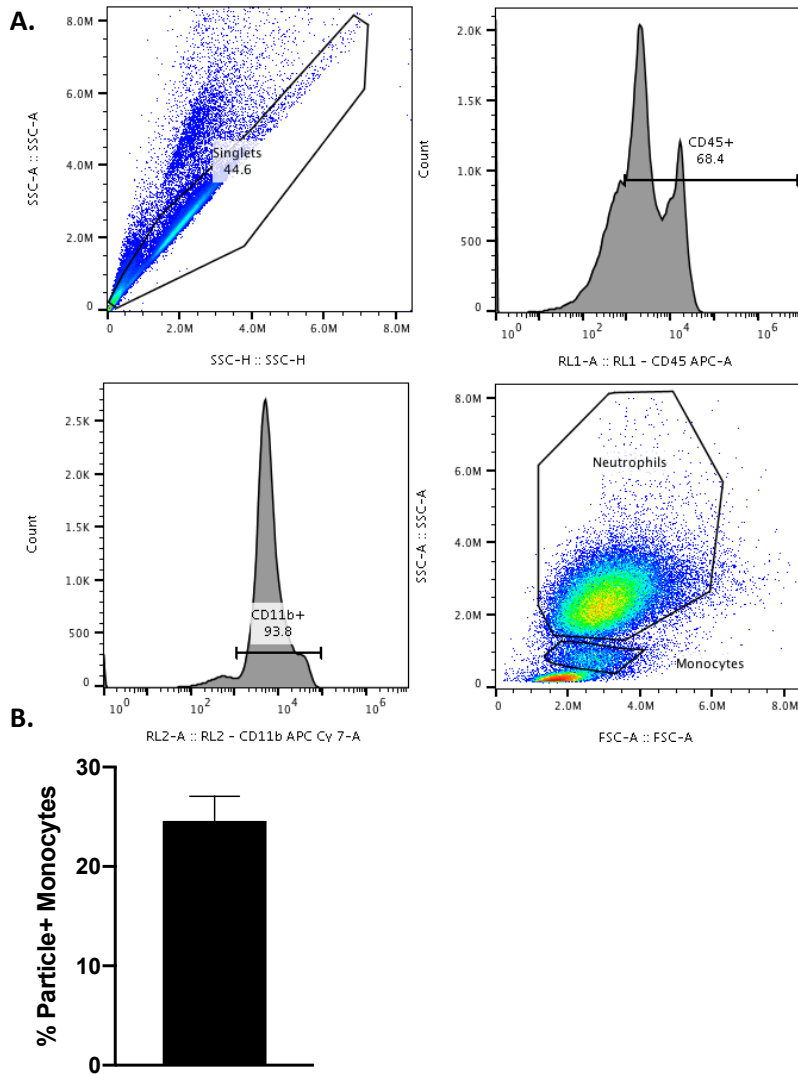
**Supplemental Figure 3. Surface Coverage versus Percent Reduction in Leukocyte Adhesion.** (a) Surface Coverage vs. % Reduction in Leukocyte Adhesion for 5  $\mu\text{m}$  particles. (b) Surface Coverage vs. % Reduction in Leukocyte Adhesion for 3  $\mu\text{m}$  particles. (c) Surface Coverage vs. % Reduction in Leukocyte Adhesion for 2  $\mu\text{m}$  particles. (d) Surface Coverage vs. % Reduction in Leukocyte Adhesion for 500 nm particles. Each data point represents at least 3 independent donors. Error bars are standard error.



**Supplemental Figure 4. Percent Reduction in Leukocyte Adhesion versus Particle Concentration for Targeted and Non-Targeted Particles**

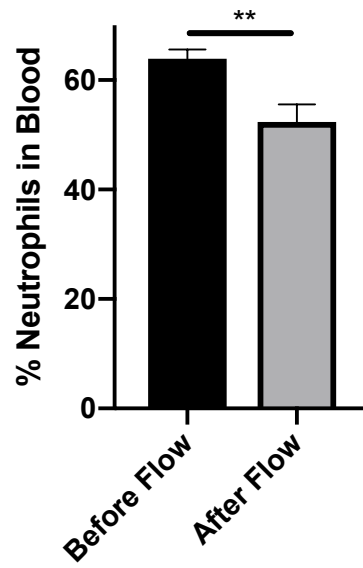


**Supplemental Figure 5. Individual trial results showing % reduction in leukocyte adhesion for 3  $\mu\text{m}$  and 5  $\mu\text{m}$  particles.**



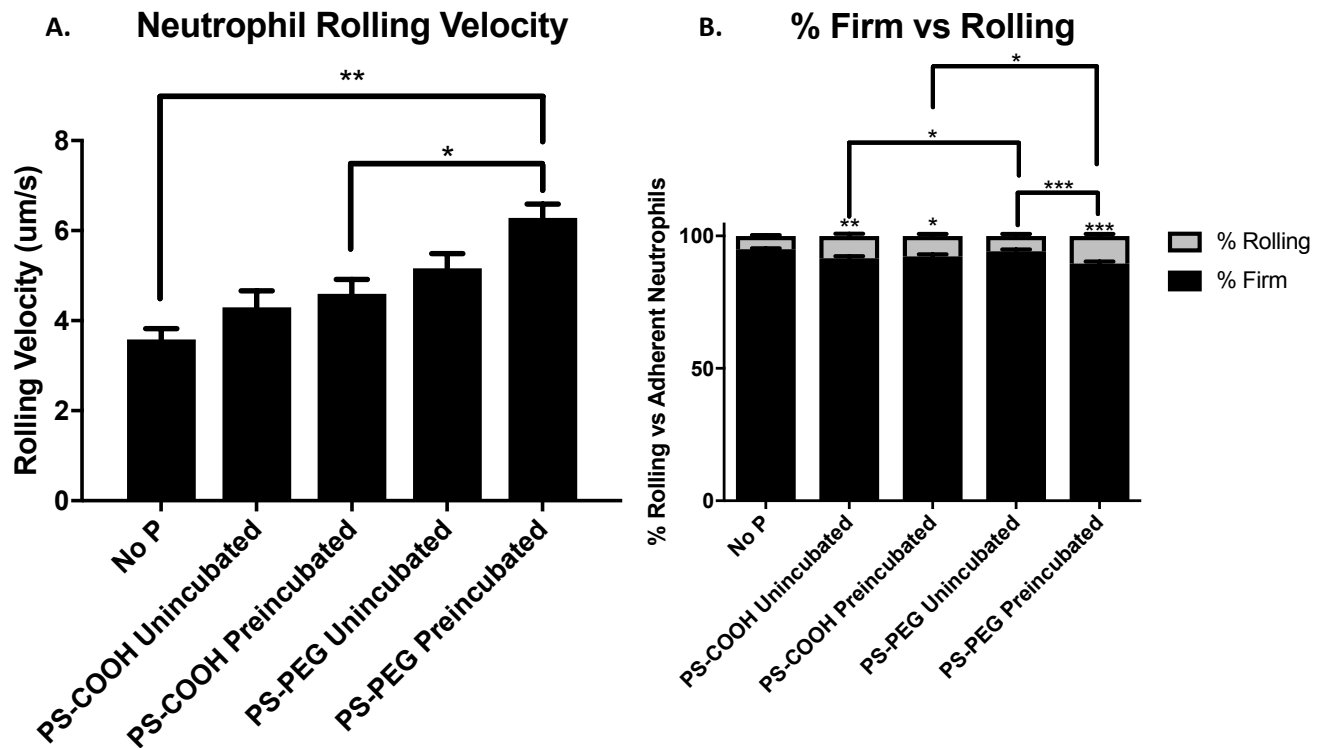
**Supplemental Figure 6. Flow Cytometry Analysis of Monocyte Particle Phagocytosis in Whole Blood *ex vivo*.** (a) Gating (right to left, top to bottom) for differentiating neutrophils from monocytes in whole blood samples. (b) Percent particle positive monocytes in whole blood after incubating for 2 hours with 2  $\mu\text{m}$  particles at a concentration of 1E7/mL. Error bars represent standard error for three separate donors, repeated in duplicate.

### % of Neutrophils in Blood Before and After Flow



Supplemental Figure 7. Flow Cytometry Analysis of Neutrophils in Whole Blood Before and After Flow Experiments.





**Supplemental Figure 8. Impact of Particle Internalization on Neutrophil Rolling Velocity and Firm Adherence in Laminar Flow.** (a) Neutrophil rolling velocity on an inflamed endothelium with and without particle internalization for 2  $\mu\text{m}$  PS-COOH and PS-PEG particles introduced at a concentration of  $1\text{E}7/\text{mL}$ , measured by tracking leukocytes through 10-second videos. (b) Percentage of firmly adherent versus rolling neutrophils with and without particle internalization for 2  $\mu\text{m}$  PS-COOH and PS-PEG particles introduced at a concentration of  $1\text{E}7/\text{mL}$ , as measured by manually counting the number of rolling versus firmly adherent neutrophils in 10-second videos. (\*) directly above a bar indicates significant difference from the untreated ("No P") condition, while other significance bars indicate significance between those two conditions ( $p < 0.05$ ). Statistical analysis was performed using one-way ANOVA using GraphPad Prism software.  $n \geq 3$  donors for each condition.