Supplemental Digital Content 1: Supplemental Methods

Isolation of microparticles

A sample of 250 µL of frozen plasma was thawed on melting ice for 1 h and centrifuged for 30 min at 18,890*g* at 20°C to pellet the microparticles. After centrifugation, 225 µL of the supernatant were removed. The pellet was resuspended in 225 µL phosphate-buffered saline (PBS) containing citrate, after which samples were centrifuged again and supernatants were removed again. The pellet containing microparticles was resuspended in 75 µL PBS-citrate for the final concentration.

Flowcytometry of microparticles

Five microliters of the microparticles suspension was diluted in 30 µL $CaCl_2$ (2.5 mmol/L)-containing PBS. Than 5 µL allophycocyanin-labelled annexin V were added to all tubes plus 5 µL of the cell-specific monoclonal antibody or isotype-matched control antibodies (total volume: 50 µL). The samples were incubated in the dark for 15 min at room temperature. After incubation, 900 µL of calcium-containing PBS were added to all tubes (except to the annexin V control, to which 900 µL of citrate-containing PBS were added). Samples were analysed for 1 min in a fluorescence automated cell sorter FACS Calibur with CellQuest software (Becton Dickinson, San Jose, CA). Both forward scatter and sideward scatter were set at logarithmic gain. To establish the origin of the microparticles, we performed a triple labeling on each sample using different fluorochromes (Fluorescein isothiocyanate (FITC)-labeled IgG_1, phycoerythrin (PE)-labeled IgG_1, and allophycocyanin conjugated annexin V). These antibodies were used to analyse the origin of the circulating microparticles: CD61-PE (exposed on thrombocytes), CD63 (expressed by activated platelets), CD14 (mostly found on macrophages), CD4 (from T-cells), CD8 (also from T-cells), CD20 (from B-cells), glycophorin A-FITC (CD235a) from erythrocytes, CD144-FITC exposed on endothelial cells,

CD62e-PE (E-selectin exposed on activated endothelial cells). Microparticles were identified based on their size and density and on their ability to bind cell-type specific CD antibodies and annexin V. Microparticles positive for annexin V, CD62e-PE and Cd144-FITC were considered to be derived from activated endothelial cells.

Antibodies

FITC-labeled IgG_1, PE-labeled $IgG_{1,}$ CD4-PE, CD8-PE, CD14-PE, CD20-FITC, CD61-PE, CD146-PE, Tissue Factor-PE were obtained from Becton Dickinson, Ig2b-PE and IgGpoly-FITC from Immuno Quality Products (Groningen, The Netherlands), CD24-PE (Serotec, Oxford, United Kingdom), CD61-FITC, CD235a from DAKO (Glostrup, Denmark), CD62e-PE from Ancell corporation (Bayport, MN), CD63-PE from Beckman Coulter Inc. (Fullerton, CA), CD144-FITC from Alexis Biochemicals (San Diego, CA), Allophycocyanin-conjugated annexinV was obtained from Caltag (Burlington, CA).