

SUPPLEMENTARY FIG. S7. Platelet-like particle (PLP) and proplatelet production in X-VIVO 10 cultures. (A) Blood platelets obtained from AllCells (top panels) and PLPs exposed to shear for 3 days after CD61-selection on day 10 (bottom panels) were stained with anti-CD41 and anti-CD62P antibodies, incubated with propidium iodide, and acquired via flow cytometry using the same settings for all samples. (B) CD34⁺ HSPCs were seeded in X-VIVO 10 with cocktail "c" (Table 1) using IL-3 from R&D Systems and maintained at 5% O_2 and pH 7.2. On day 5, cells were washed, resuspended at pH 7.4/20% O_2 , and supplemented with cocktail 3. On day 7, Mks were isolated using a CD61-selection column (>80% CD41⁺), supplemented with 100 ng/mL Tpo and 100 ng/mL SCF, and plated on fibrinogen-coated glass slides. On day 12, cells were fixed and stained for β-tubulin and imaged for proplatelet formation. Scale bar = 50 μm (C) CD34⁺ HSPCs were seeded in X-VIVO 10 with 100 ng/mL Tpo and 100 ng/mL SCF. On day 5, 6.25 mM NIC was added. At day 9, proplatelets were imaged with phase-contrast microscopy. Scale bar = 20 μm.