

Supplementary Figure E1. Induction of CD36E cells with hemin, DMSO, or PMA. CD36E cells were cultured in the serum-free expansion medium containing hemin, DMSO, or PMA at indicated concentrations. (A) Cell proliferation was measured in triplicate by Trypan blue staining at indicated time points. DMSO (1.25% and 2%) and 50 μM hemin were cytotoxic to CD36E cells, leading to rapid cell death, while 25 μM hemin was less toxic. PMA (10 ng/mL and 50 ng/mL) also inhibited CD36E proliferation but was less toxic compared with DMSO and hemin. (B) Day 3 cells cultured with 25 μM hemin or 10 ng/mL PMA were stained with Wright-Giemsa and analyzed by microscopy at $63 \times$ magnification, showing cells were filled with vacuoles. Scale bars: 10 μm. (C) Whole cell lysates were prepared from hemin- (25 μM) or PMA- (10 ng) treated CD36E cells on days 4 and 5, followed by immunoblotting using antibodies against globin chains (α, β, and γ) and glyceraldehyde phosphate dehydrogenase (an internal control). The results showed a substantial decrease in β-chain expression in the cells as compared with cells at day 0, but no such difference was observed for α- and γ-chain expression. Immunoblotting was not performed on cells treated with DMSO, due to the rapid cell death upon treatment.

Supplementary Table E1. Fold changes of cell populations in various cytokines and growth factors

	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8	Day 9
EPO	0.94 ± 0.12	0.54 ± 0.12	0.55 ± 0.18	1.00 ± 0.47	0.08 ± 0.06	0.00 ± 0.01			
SCF+IL-3	1.23 ± 0.09	0.71 ± 0.03	0.51 ± 0.06	0.38 ± 0.04	0.00 ± 0.01	0.00 ± 0.00			
SCF	0.99 ± 0.08	0.78 ± 0.04	0.54 ± 0.03	0.21 ± 0.06	0.02 ± 0.02	0.00 ± 0.00			
EPO+SCF	0.97 ± 0.05	1.48 ± 0.26	2.44 ± 0.12	2.82 ± 0.42	3.12 ± 0.37	5.47 ± 0.79	8.00 ± 1.32	11.17 ± 0.32	12.42 ± 0.38
IL-3	0.79 ± 0.11	0.55 ± 0.11	0.43 ± 0.05	0.27 ± 0.08	0.00 ± 0.01	0.01 ± 0.02			
EPO+IL-3	0.89 ± 0.11	0.66 ± 0.10	0.44 ± 0.06	0.21 ± 0.04	0.04 ± 0.01	0.00 ± 0.00			
TPO	0.94 ± 0.18	0.57 ± 0.13	0.42 ± 0.01	0.27 ± 0.12	0.03 ± 0.03	0.00 ± 0.00			
EPO+TPO	0.82 ± 0.08	0.61 ± 0.08	0.37 ± 0.13	0.34 ± 0.13	0.02 ± 0.02	0.01 ± 0.02			
EPO+SCF+IL-3	1.18 ± 0.11	1.73 ± 0.30	2.96 ± 0.39	3.39 ± 0.48	4.44 ± 0.86	5.48 ± 0.53	7.82 ± 0.86	10.75 ± 2.09	11.54 ± 0.83
None	0.67 ± 0.20	0.58 ± 0.06	0.27 ± 0.03	0.33 ± 0.04	0.04 ± 0.03	0.00 ± 0.00			

Values are mean \pm SD (n = 3).

Supplementary Table E2. Fold changes of cell populations in different cytokine concentrations

	Day 1	Day 2	Day 3	Day 4	Day 5
3 IU EPO/mL	1.33 ± 0.12	0.80 ± 0.28	0.42 ± 0.13	0.32 ± 0.15	0.23 ± 0.03
9 IU EPO/mL	0.87 ± 0.26	0.50 ± 0.06	0.27 ± 0.23	0.47 ± 0.09	0.36 ± 0.08
27 IU EPO/mL	0.87 ± 0.47	0.64 ± 0.19	0.18 ± 0.08	0.34 ± 0.08	0.17 ± 0.13
100 ng SCF/mL	0.99 ± 0.18	1.10 ± 0.10	0.29 ± 0.10	0.29 ± 0.05	0.22 ± 0.05
300 ng SCF/mL	1.25 ± 0.31	0.68 ± 0.12	0.46 ± 0.13	0.29 ± 0.17	0.22 ± 0.07
900 ng SCF/mL	1.17 ± 0.23	0.84 ± 0.39	0.21 ± 0.08	0.30 ± 0.06	0.60 ± 0.72
3 IU EPO/mL+ 100 ng SCF/mL	1.90 ± 0.12	1.83 ± 0.26	1.81 ± 0.46	2.08 ± 0.13	2.34 ± 0.95
9 IU EPO/mL + 300 ng SCF/mL	1.69 ± 0.08	1.88 ± 0.40	1.18 ± 0.32	1.43 ± 0.20	1.87 ± 0.30
27 IU EPO/mL + 900 ng SCF/mL	1.21 ± 0.24	1.47 ± 0.20	1.37 ± 0.47	1.30 ± 0.27	1.72 ± 0.55

Values are mean \pm SD (n = 3).

Supplementary Table E3. Fold changes of cell populations in different mitogens

	Day 1	Day 2	Day 3	Day 4	Day 5
25 μM Hemin	1.27 ± 0.35	0.84 ± 0.19	0.84 ± 0.07	0.84 ± 0.07	0.57 ± 0.30
50 μM Hemin	1.21 ± 0.12	0.71 ± 0.05	0.54 ± 0.08	0.44 ± 0.16	0.23 ± 0.03
1.25% DMSO	1.58 ± 0.23	1.14 ± 0.19	0.28 ± 0.10	0.34 ± 0.05	0.30 ± 0.23
2% DMSO	1.29 ± 0.29	0.79 ± 0.05	0.59 ± 0.05	0.39 ± 0.13	0.48 ± 0.57
10 ng PMA/mL	1.46 ± 0.14	1.33 ± 0.09	1.11 ± 0.02	1.25 ± 0.12	0.99 ± 0.22
50 ng PMA/mL	1.39 ± 0.34	1.09 ± 0.12	1.16 ± 0.16	1.12 ± 0.13	0.79 ± 0.86

Values are mean \pm SD (n = 3).