Supplemental Table S1: Donor Information. Table includes identifying number, age, sex, SCD genotype, hydroxyurea (HU) usage, history of recent transfusion therapy, percentage of sickle hemoglobin (HbS), fetal hemoglobin (HbF), normal adult hemoglobin (HbA), hemoglobin A2 (HbA2), hemoglobin C (HbC), white blood cell count (WBC), hemoglobin (HGB), hematocrit (HCT), platelet count (PLT), mean corpuscular volume (MCV), mean corpuscular hemoglobin concentration (MCHC), and red blood cell distribution width (RDW). * Indicates pediatric donors.

	SCD Donors (n=13)	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9***	**10**	**11***	**12**	**13***
Donor History and Demographics	**Age (years)** Range: 7-39	31	39	28	21	27	21	23	29	13	31	14	29	7
	Sex 8M, 5F	M	M	F	F	M	F	F	F	M	M	M	M	M
	Genotype 11 HbSS, 2 HbSC	SS	SS	SS	SS	SC	SS	SS	SC	SS	SS	SS	SS	SS
	HU Usage 13 Yes, 0 No	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
	Recent Transfusion 3 Yes, 10 No	No	Yes	Yes	No	No	No	No	No	No	No	No	Yes	No
Hemoglobin Variant Analysis	**HbS (%)** Range: 48.4-89.2	78.4	64.5	56.5	78.3	48.4	78.6	84.9	49.6	89.2	81.6	84.7	61.9	77.1
	HbF (%) Range: 6.4-26.5	18.8	19.9	26.5	18.9	6.4	18.4	11.8	9.3	7.4	14.9	12.7	17.0	19.9
	HbA (%) Range: 0.0-18.7	0.0	13.4	13.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	18.7	0.0
	HbA2 (%) Range: 2.2-3.7	2.8	2.2	3.7	2.8	3.8	3.0	3.3	3.2	3.4	3.5	2.6	2.4	3.0
	HbC (%) Range: 0.0-41.4	0.0	0.0	0.0	0.0	41.4	0.0	0.0	38.0	0.0	0.0	0.0	0.0	0.0
Complete Blood Count Analysis	**WBC (k*10^9/µL)** Range: 4.00-12.02	8.54	8.35	4.00	9.75	4.52	6.53	9.28	4.96	11.10	5.30	8.10	8.10	12.02
	HGB (g/dL) Range: 6.5-10.0	6.5	7.2	10.0	8.4	9.3	8.9	6.8	9.7	9.8	9.5	8.9	9.1	8.4
	HCT (%) Range: 18.2-27.8	18.2	21.6	27.7	23.1	24.5	24.2	20.6	27.3	27.8	27.5	24.0	24.3	22.8
	PLT (k*10^9/µL) Range: 61-676	446	335	122	539	61	500	381	196	460	277	365	331	676
	MCV (fL) Range: 80.8-115.9	115.9	107.5	102.2	84.6	84.5	86.1	84.4	80.8	96.0	93.0	102.0	103.0	92.7
	MCHC (g/dL) Range: 33.0-38.0	35.7	33.3	36.1	36.4	38.0	36.8	33.0	35.5	35.3	34.5	37.7	37.4	36.8
	RDW (%) Range: 14.5-21.6	18.5	18.9	15.6	20.5	17.2	17.4	18.9	14.5	17.8	16.9	19.9	21.6	14.7

Supplemental Table S2: RNA sequencing quality control assessment. Table includes RNA sample ID, preconditioning treatment, and various quality control results (nucleic acid concentration, total RNA quantity, purity ratio, RNA integrity number (RIN), and DV200). RNA quality/integrity was deemed excellent via Tape Station analysis, which allowed for downstream analysis via RNA-sequencing. Note that samples P1, C1, and C2 did not produce a RIN score due to low input sample concentration, however review of the 18S and 28S peaks indicated good quality.

ID	**Preconditioning Treatment** (oxygen level)	**Nanodrop**			**Qubit**		**Tape Station**	
		Nucleic Acids (ng/µL)	**Total RNA** (ng)	**260/ 280** Ratio	**Nucleic Acids** (ng/µL)	**Total RNA** (ng)	**RIN** (#)	**DV200** (%)
A1	21%	28.9	404.6	1.93	35.8	501.2	9.8	80.46
A2	21%	38.9	544.6	1.95	49.4	691.6	10	84.73
A3	21%	98.4	1377.6	1.96	64.2	898.8	10	91.85
P1	12%	18.4	257.6	1.69	14.2	198.8	-	74.54
P2	12%	19.4	271.6	1.9	23.2	324.8	10	78.32
P3	12%	28.8	403.2	1.91	36.2	506.8	9.8	76.52
C1	Cyclic (12%-5%)	16.2	226.8	1.96	11.1	155.4	-	76.83
C2	Cyclic (12%-5%)	19.5	273.0	1.61	8.5	119.3	-	70.78
C3	Cyclic (12%-5%)	26.8	375.2	1.94	18.5	259.0	10	76.68
S1	5%	30.8	431.2	1.88	18.2	254.8	10	76.01
S2	5%	38.5	269.5	1.9	25.0	175.0	10	86.09
S3	5%	40.7	569.8	1.81	28.6	400.4	10	81.80
Avg	Average	33.8	450.4	1.87	27.7	373.8	9.96	79.55
SD	Standard Deviation	22.0	312.8	0.11	16.4	238.1	0.09	5.78

Supplemental Table S3: Top Gene Ontologies, 5% vs. 21%. Table includes the top 25 GO pathways for the 5% to 21% preconditioning comparison, sorted by adjusted p-value. Also included are the number of DEGs for each, pathway percentage, and p-adj.

Gene Ontology Pathway	# DEG	% Pathway	P-adj
Muscle filament sliding	5	13.16	3.82E-08
Cardiac muscle contraction	4	9.09	6.54E-06
Striated muscle contraction	3	23.08	2.10E-05
Regulation of the force of heart contraction	3	16.67	3.73E-05
Ventricular cardiac muscle tissue morphogenesis	3	12.00	7.33E-05
Sarcomere organization	3	11.11	7.56E-05
Cerebral cortex tangential migration	2	50.00	0.00039
Cell adhesion	5	1.11	0.00065
Cardiac muscle fiber development	2	28.57	0.00065
Positive regulation of cation channel activity	2	28.57	0.00065
Limb bud formation	2	20.00	0.00108
Platelet degranulation	3	2.97	0.00141
Muscle contraction	3	2.97	0.00141
Adult heart development	2	15.39	0.00141
Central nervous system development	3	2.56	0.00201
Artery morphogenesis	2	9.52	0.00284
Positive regulation of G1/S transition of mitotic cell cycle	2	9.09	0.00291
Axon guidance	3	1.92	0.00365
Peptidyl-tyrosine phosphorylation	3	1.95	0.00365
Regulation of heart rate	2	6.45	0.00393
Cell communication	2	6.90	0.00393
Long-term memory	2	6.90	0.00393
Regulation of heart contraction	2	6.67	0.00393
ATP metabolic process	2	6.45	0.00393
In utero embryonic development	3	1.67	0.00419

Supplemental Table S4: Top Gene Ontologies, 5% vs. 12%. Table includes the top 25 GO pathways for the 5% to 12% preconditioning comparison, sorted by adjusted p-value. Also included are the number of DEGs for each, pathway percentage, and p-adj.

Gene Ontology Pathway	# DEG	% Pathway	P-adj
Cardiac muscle contraction	7	15.91	5.85E-11
Muscle filament sliding	5	13.16	2.22E-08
Regulation of the force of heart contraction	4	22.22	1.81E-07
Regulation of heart rate	4	12.90	9.68E-07
Cardiac myofibril assembly	3	25.00	1.06E-05
Striated muscle contraction	3	23.08	1.08E-05
Response to muscle stretch	3	18.75	1.61E-05
Platelet degranulation	4	3.96	3.79E-05
Muscle contraction	4	3.96	3.79E-05
Sarcomere organization	3	11.11	4.68E-05
Skeletal muscle thin filament assembly	2	40.00	0.00031
Positive regulation of the force of heart contraction	2	40.00	0.00031
Regulation of cardiac muscle contraction by calcium ion signaling	2	28.57	0.00042
Cardiac muscle fiber development	2	28.57	0.00042
Cellular response to caffeine	2	28.57	0.00042
Cardiac muscle hypertrophy	2	25.00	0.00050
Cell-matrix adhesion	3	3.30	0.00088
Adult heart development	2	15.39	0.00088
Cardiac muscle tissue morphogenesis	2	16.67	0.00088
Calcium ion transport into cytosol	2	15.39	0.00088
Cell communication by electrical coupling	2	15.39	0.00088
Neuron migration	3	3.00	0.00090
Regulation of cardiac muscle contraction by regulation of the release of sequestered calcium ion	2	12.50	0.00112
Response to pain	2	12.50	0.00112
Positive regulation of protein phosphorylation	3	2.42	0.00147

Supplemental Table S5: Top Gene Ontologies, Cyclic vs. 21%. Table includes the top 25 GO pathways for the cyclic to 21% preconditioning comparison, sorted by adjusted p-value. Also included are the number of DEGs for each, pathway percentage, and p-adj.

Gene Ontology Pathway	# DEG	% Pathway	P-adj
Cell adhesion	31	6.86	0
Signal transduction	41	3.82	0
Nervous system development	22	7.07	0
Extracellular matrix organization	23	11.98	0
Positive regulation of transcription from RNA polymerase II promoter	36	3.74	0
Cardiac muscle contraction	16	36.36	0
Muscle filament sliding	12	31.58	2.21E-11
Axon guidance	21	13.46	2.50E-10
Chemical synaptic transmission	19	7.82	3.78E-10
Neuron migration	14	14.00	4.41E-10
Platelet degranulation	14	13.86	4.41E-10
Heart development	17	9.88	4.41E-10
Positive regulation of synapse assembly	12	19.67	4.41E-10
Ventricular cardiac muscle tissue morphogenesis	9	36.00	2.42E-09
Central nervous system development	14	11.97	2.56E-09
Regulation of cardiac conduction	11	19.64	3.09E-09
Regulation of heart contraction	9	30.00	7.75E-09
Positive regulation of cell proliferation	23	5.07	1.20E-08
Nucleosome assembly	12	13.79	1.27E-08
Skeletal system development	14	10.22	1.30E-08
Learning	10	18.18	3.79E-08
Negative regulation of endopeptidase activity	13	10.48	4.07E-08
Muscle contraction	12	11.88	4.99E-08
Retinoid metabolic process	10	16.95	6.11E-08
Regulation of the force of heart contraction	7	38.89	1.67E-07

Supplemental Table S6: Top Gene Ontologies, Cyclic vs. 12%. Table includes the top 25 GO pathways for the cyclic to 12% preconditioning comparison, sorted by adjusted p-value. Also included are the number of DEGs for each, pathway percentage, and p-adj.

Gene Ontology Pathway	# DEG	% Pathway	P-adj
Muscle filament sliding	12	31.58	0
Cardiac muscle contraction	14	31.82	0
Platelet degranulation	14	13.86	1.54E-10
Cell adhesion	18	3.98	1.54E-10
Ventricular cardiac muscle tissue morphogenesis	8	32.00	1.85E-10
Sarcomere organization	8	29.63	2.50E-10
Negative regulation of endopeptidase activity	12	9.68	2.73E-10
Axon guidance	13	8.33	3.66E-10
Regulation of heart contraction	8	26.67	3.66E-10
Extracellular matrix organization	16	8.33	3.66E-10
Muscle contraction	10	9.90	3.60E-09
Cardiac myofibril assembly	6	50.00	7.81E-09
Regulation of heart rate	7	22.58	1.74E-08
Regulation of cardiac conduction	8	14.29	1.96E-08
Retinoid metabolic process	8	13.56	2.68E-08
Regulation of heart rate by cardiac conduction	7	20.00	2.99E-08
Skeletal system development	10	7.30	3.70E-08
Regulation of the force of heart contraction	6	33.33	3.70E-08
Heart development	10	5.81	2.64E-07
Regulation of cardiac muscle contraction by regulation of the release of sequestered calcium ion	5	31.25	1.33E-06
Lipoprotein metabolic process	6	15.79	1.56E-06
Response to hypoxia	9	5.70	1.68E-06
Membrane depolarization during SA node cell action potential	4	80.00	1.94E-06
Positive regulation of ERK1 and ERK2 cascade	9	5.20	3.24E-06
Fibrinolysis	5	23.81	3.39E-06