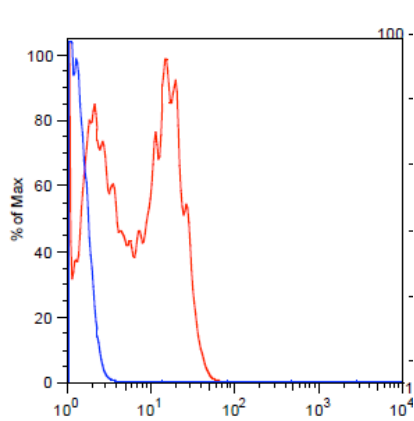


SUPPLEMENTARY DATA FOR:

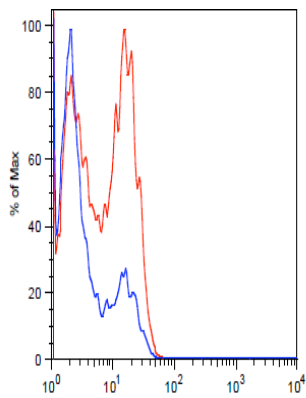
Studies on the Antiproliferative Effects of Tropolone Derivatives in Jurkat T-Lymphocyte Cells

Sophia N. Ononye, Michael D. Van Heyst, Charles Giardina, Dennis L. Wright and Amy C. Anderson

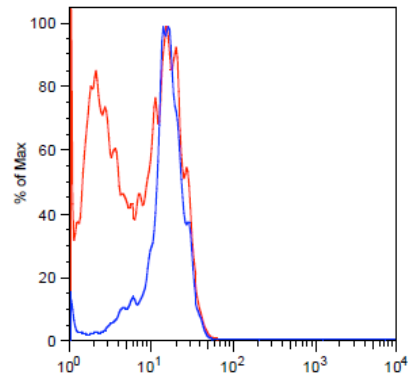
Department of Pharmaceutical Sciences, University of Connecticut, 69 N. Eagleville Rd., Storrs, CT 06269



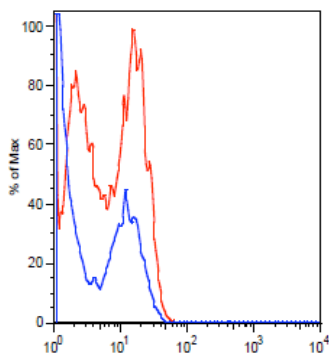
a) Experimental/compensation controls [2° antibody (blue); untreated control (red)]



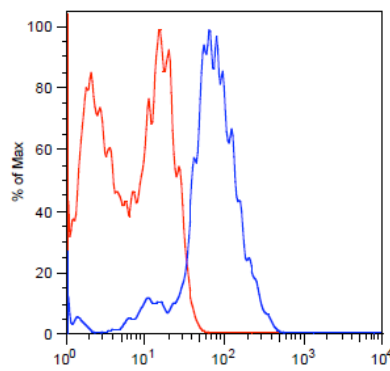
b) Compound 4



c) Compound 1

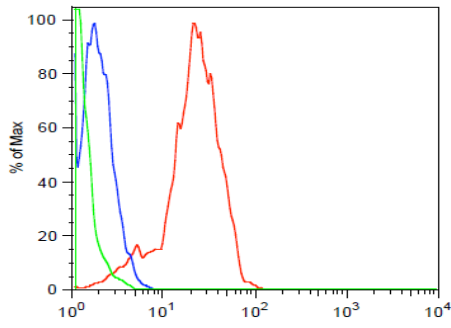


d) Compound 2

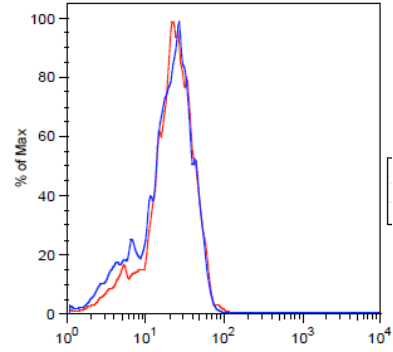


e) Vorinostat

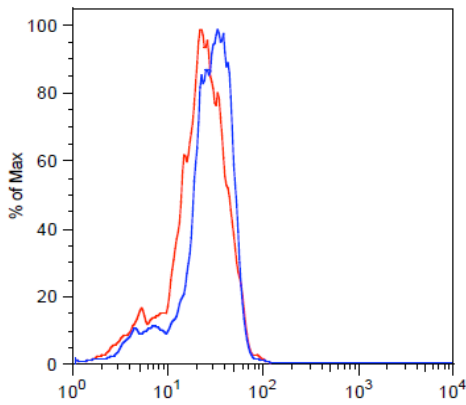
Figure S1a. Modulation of histone H3K9Ac antibodies in Jurkat cells after a 12h treatment with 10 μ M HDACi. **Note:** Untreated control for pertinent HDACi treatments represented in red and HDACi represented in blue on the corresponding histograms.



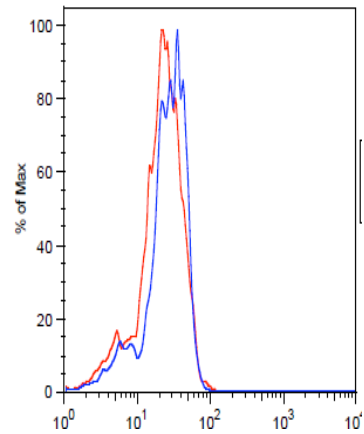
Experimental/compensation controls [unlabeled cells (green); 2° antibody (blue); untreated control (red)].



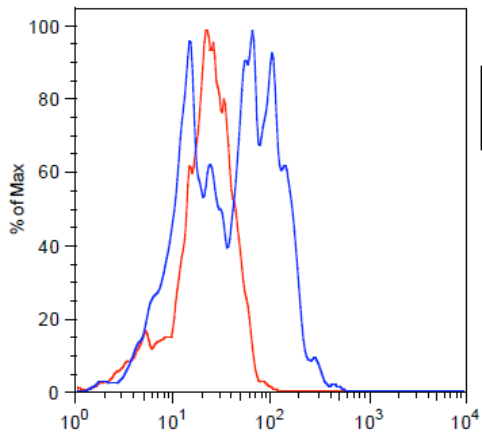
Compound 4



Compound 1

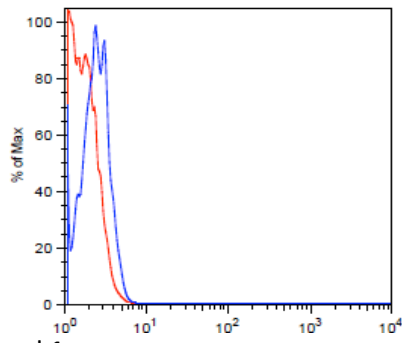


Compound 2

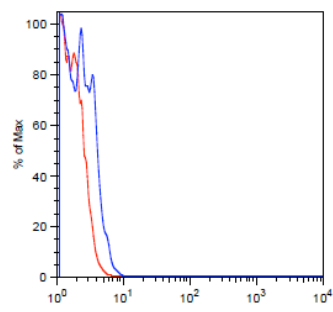


Vorinostat

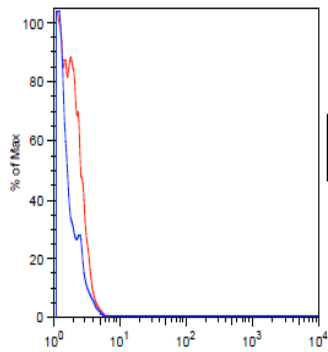
Figure S1b. Modulation of histone H3K23Ac antibodies in Jurkat cells after a 12h treatment with 10 μ M HDACi. **Note:** Untreated control for pertinent HDACi treatments represented in red and HDACi represented in blue on the corresponding histograms.



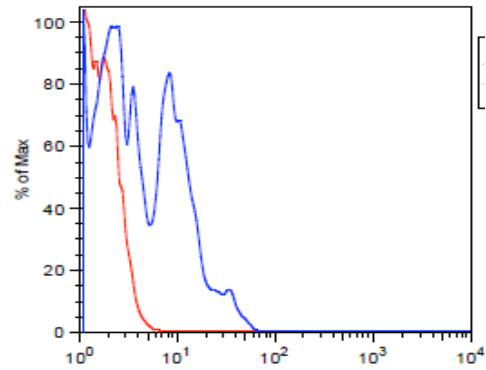
Compound 1



Compound 2



Compound 4



Vorinostat

Figure S2. Evaluation of p21 expression in Jurkat cells after a **24h** treatment with **10 μ M** HDACi. **Note:** Untreated control for pertinent HDACi treatments represented in red and HDACi represented in blue on the corresponding histograms.

Table S1. Time-dependent antiproliferative effects of tropolones and vorinostat on cell cycle progression

Time-dependent analysis of the antiproliferative effects of tropolones on cell cycle progression as measured via FACS analysis in Jurkat cells after a 24h treatment.				
Treatment	24h			
	%<G0/G1	%G1	%S	%G2-M
Control	2.39	60.90	20.90	14.30
10 μ M Vorinostat	48.90	7.46	19.90	12.30
10 μ M Compound 1	13.90	70.20	13.30	1.81
10 μ M Compound 2	13.90	66.50	15.80	2.58
10 μ M Compound 4	1.62	63.00	20.60	13.50

Time-dependent analysis of the antiproliferative effects of tropolones on cell cycle progression as measured via FACS analysis in Jurkat cells after a 36h treatment.				
Treatment	36h			
	%<G0/G1	%G1	%S	%G2-M
Control	1.80	59.10	23.00	14.20
10 μ M Vorinostat	60.80	6.90	15.60	14.90
10 μ M Compound 1	18.50	64.20	13.30	2.97
10 μ M Compound 2	13.40	65.90	14.60	4.35
10 μ M Compound 4	1.22	60.70	21.80	14.60

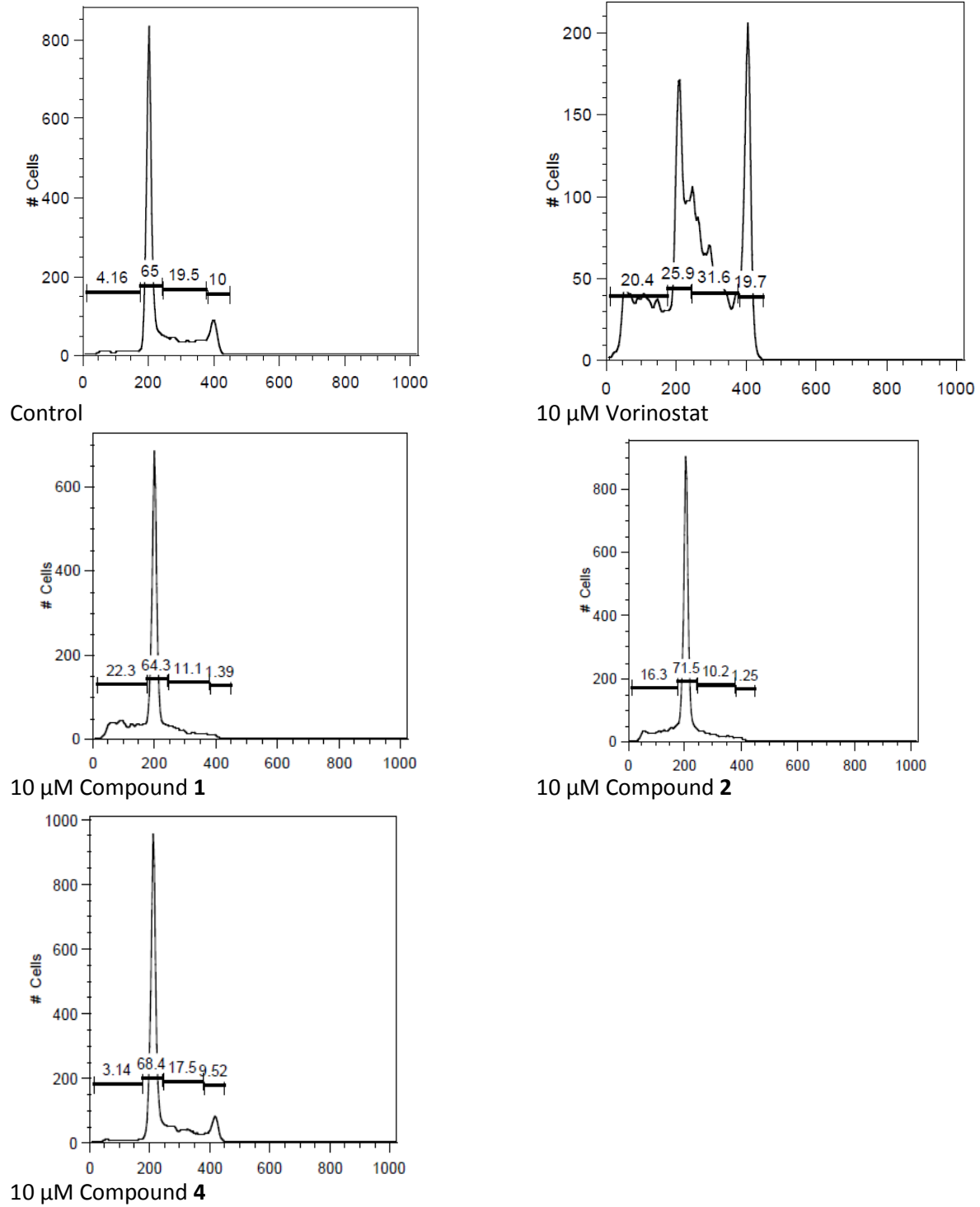
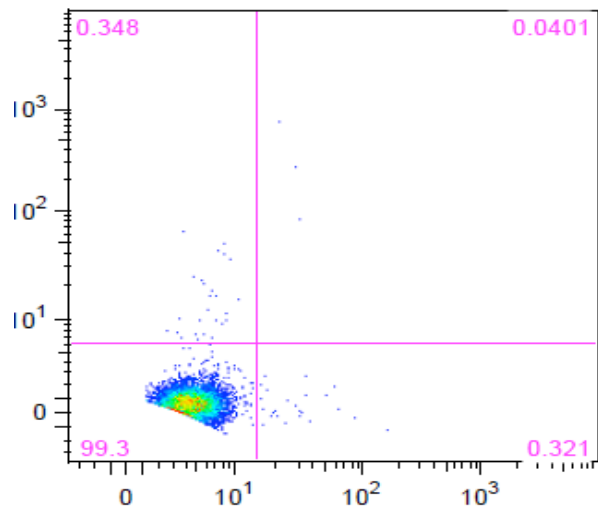
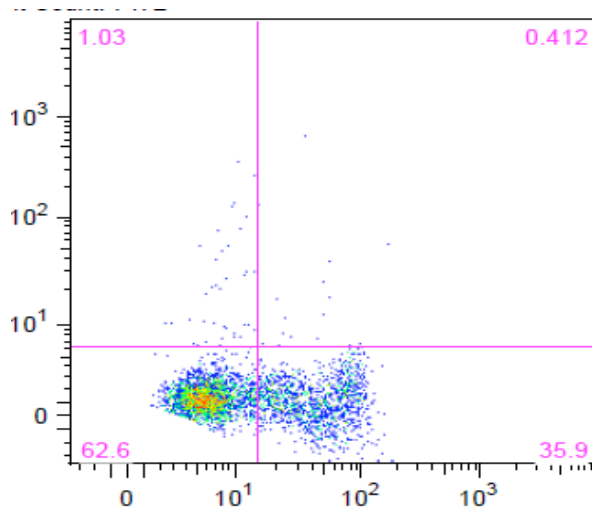


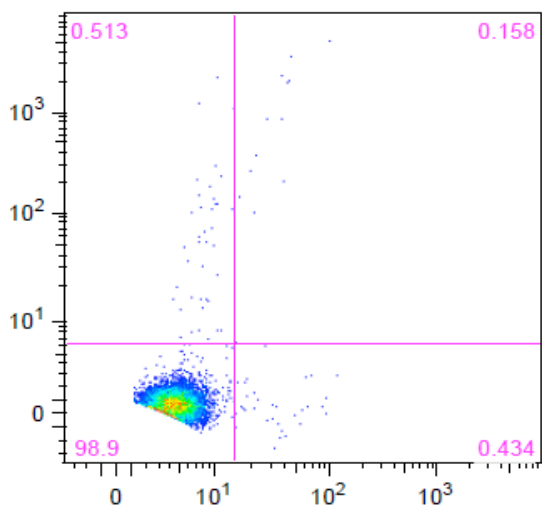
Figure S3. Histograms for cell cycle analysis



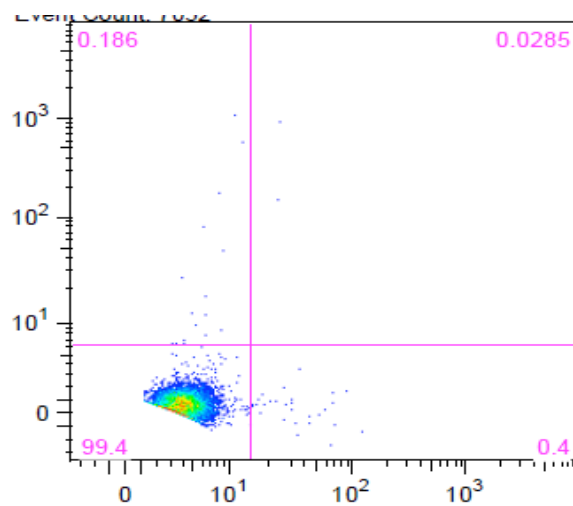
a) Control



b) Vorinostat



c) Compound 1



d) Compound 2

Figure S4: Evaluation of Caspase 8 activation in Jurkat cells after a 24h treatment with 10 μ M HDACi: y axis represents PI response whereas x axis represents caspase-8-carboxyfluorescein (FAM) response. Quadrant 1 (bottom left) represents intact (live) cells (FAM⁻, PI⁻); Quadrant 2 (bottom right) represents caspases-8 responsive cells (FAM⁺, PI⁻).

Table S2. Time-dependent analysis of caspase-3/7 activation in Jurkat cells

Treatment	Caspase-3/7 Activity (Mean RLU)			
	6h	12h	24h	48h
Control	59,417	69,935	136,627	109,510
Vorinostat	53,518	157,988	125,484	95,463
Compound 1	88,661	97,513	76,948	105,357
Compound 2	73,949	110,650	80,438	71,048

Table S3. Standard deviation values for caspase-3/7 analysis in Jurkat cells

Treatment	6h	12h	24h	48h
Control	10002.74	7003.63	31846.96	4710.453
Vorinostat	7939.778	8869.66	20509.97	10357.53
Compound 1	11860.49	6312.14	5292.832	7390.019
Compound 2	21813.73	10670.77	8117.586	6650.594