

SUPPORTING INFORMATION

Discovery of a fluorescent probe with HDAC6 selective inhibition

Yingjie Zhang^{1,2*}, Jin Yan³, Tso-Pang Yao^{3*}

¹ Department of Medicinal Chemistry, School of Pharmaceutical Sciences, Shandong University, Ji'nan, Shandong, 250012, P.R. China

² Key Laboratory of Chemical Biology (Ministry of Education), School of Pharmaceutical Sciences, Shandong University, Ji'nan, Shandong, 250012, P.R. China

³ Department of Pharmacology and Cancer Biology, Duke University, Durham, North Carolina, 27710, U.S.A.

Correspondence should be addressed to Y. Z. (email: zhangyingjie@sdu.edu.cn) or T.-P. Y. (email: tsopang.yao@duke.edu)

Contents

1. HDACs isoform selectivity comparison of 6b and tubastatin A.	S2
2. ¹ H NMR and ¹³ C NMR spectra of 6a	S3
3. HPLC analysis of 6a	S4
4. ¹ H NMR and ¹³ C NMR spectra of 6b	S5
5. HPLC analysis of 6b	S6

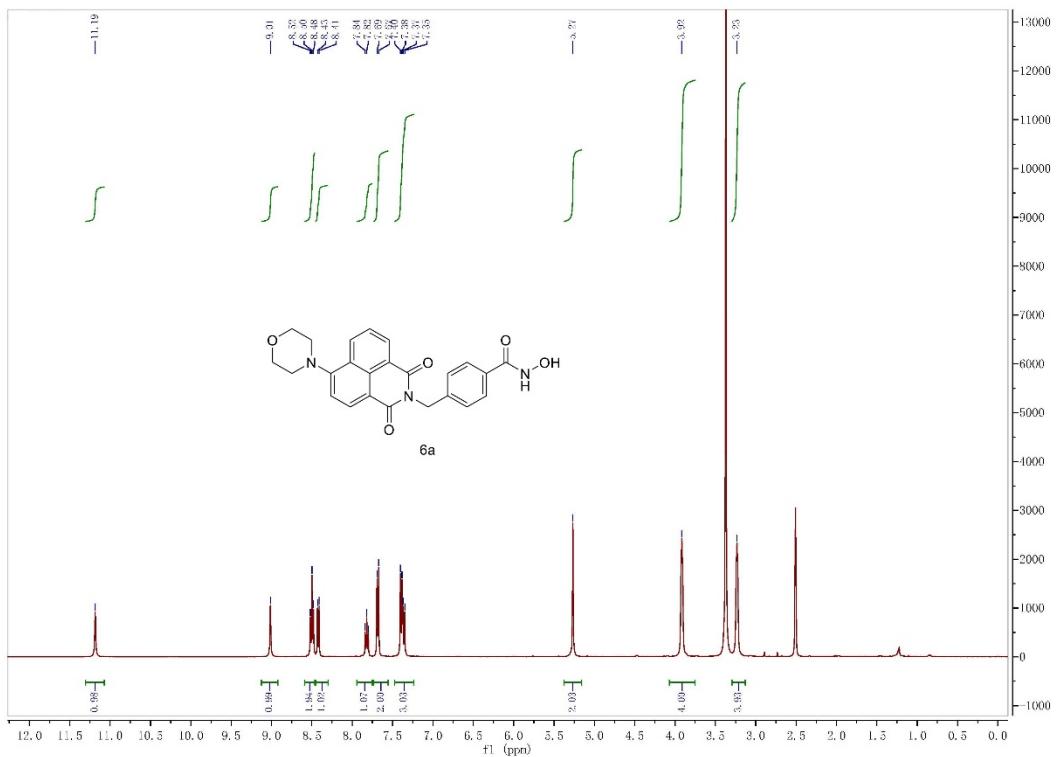
Table S1. HDACs isoform selectivity comparison of **6b** and tubastatin A.

	6b	tubastatin A
	IC ₅₀ (nM) ^a	IC ₅₀ (nM) ^b
HDAC1	>20000	16400±2600
HDAC2	>100000	>30000
HDAC3	>30000	>30000
HDAC4	>100000	>30000
HDAC5	>100000	>30000
HDAC6	139.0±1.4	15±1
HDAC7	>100000	>30000
HDAC8	3050±210	854±40
HDAC9	>30000	>30000
HDAC10	1870±50	>30000
HDAC11	>10000	>30000

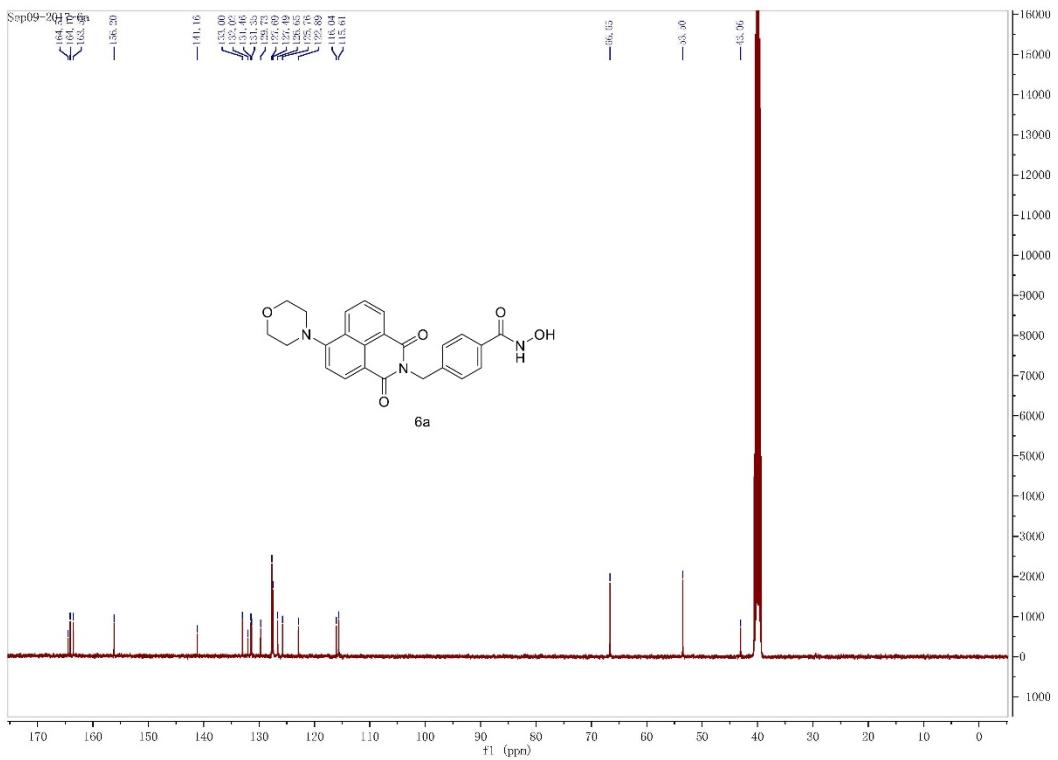
^a Assays were performed in replicate (n = 3); IC₅₀ values are shown as mean ± SD.

^b Cited from K. V. Butler, J. Kalin, C. Brochier, G. Vistoli, B. Langley, A. P. Kozikowski, Rational design and simple chemistry yield a superior, neuroprotective HDAC6 inhibitor, tubastatin A. *J. Am. Chem. Soc.* 132 (2010), 10842-10846.

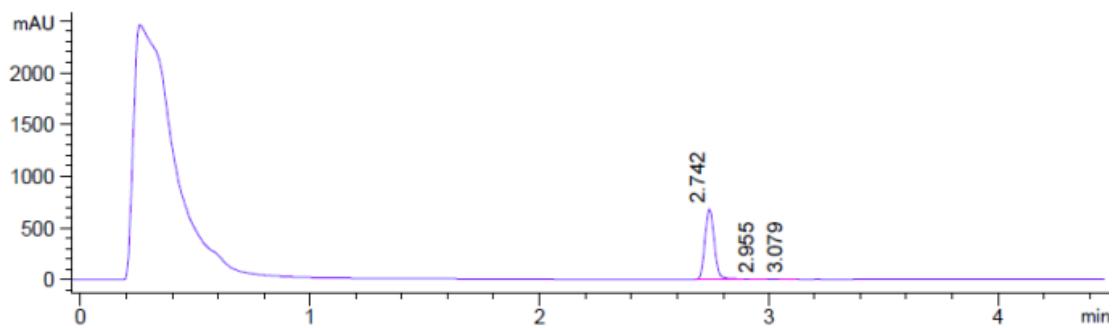
¹H NMR of **6a**



¹³C NMR of **6a**

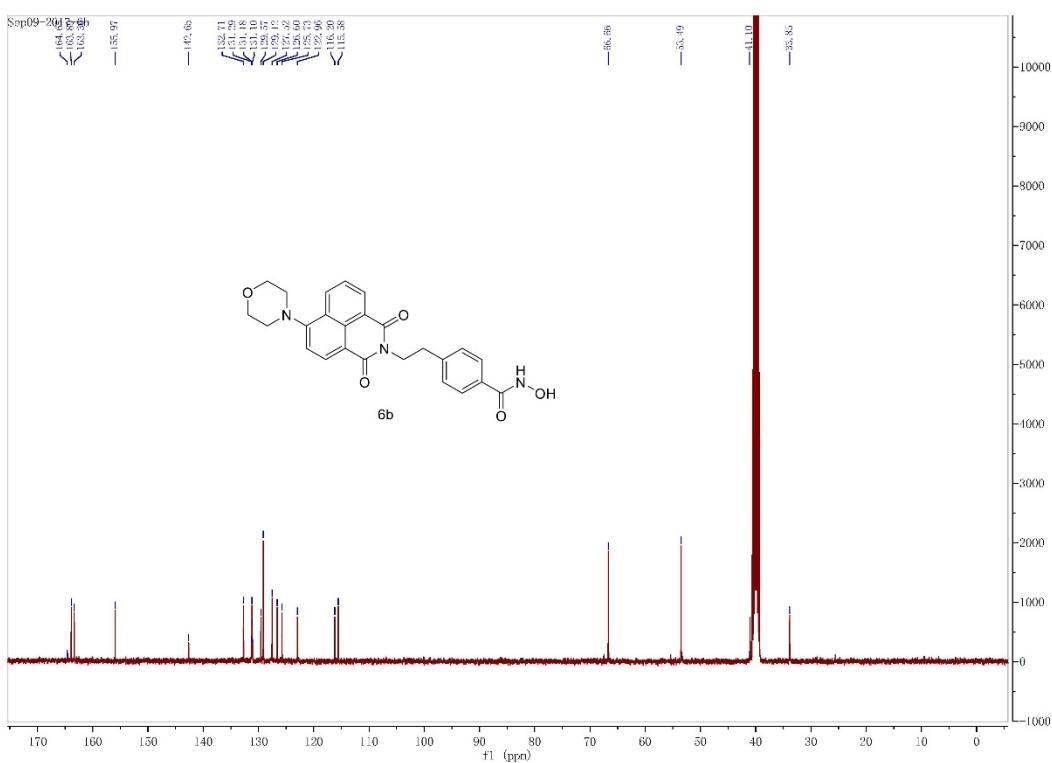
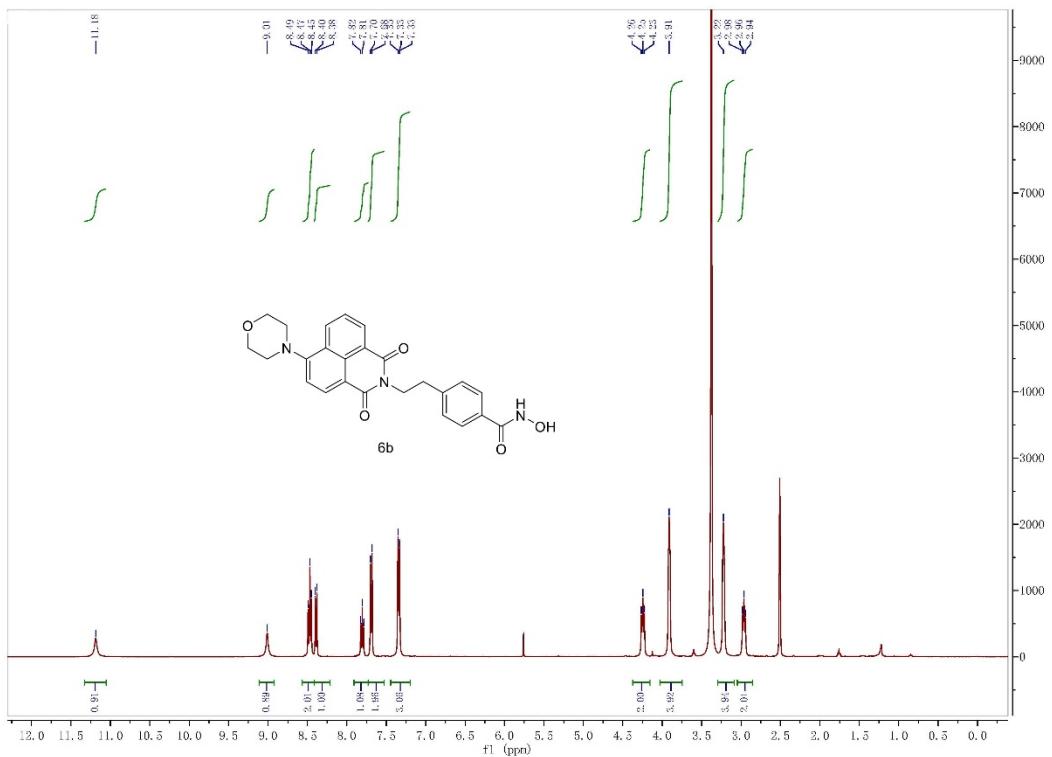


HPLC analysis of **6a**

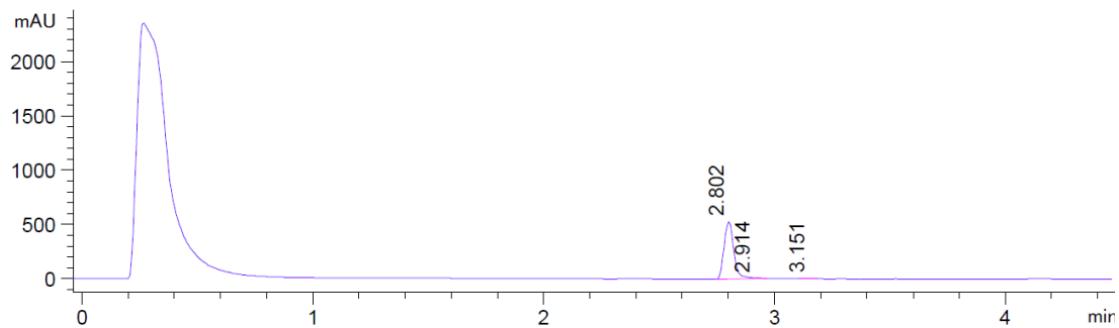


Instrument	Agilent 1200						
Column	Luna-C18(2) 2.0*50mm, 5μm						
Mobile phase A (MPA)	H ₂ O+0.037 % (v/v) TFA						
Mobile phase B (MPB)	ACN+0.018 % (v/v) TFA						
Flow rate	0.8 mL/min						
Gradient Ratio	Time (min)	0.00	0.40	3.40	3.85	3.86	4.50
	MPA (%)	99	99	10	0	99	99
	MPB (%)	1	1	90	100	1	1
Detection	220 nm						

¹H NMR of **6b**



HPLC analysis of **6b**



Instrument	Agilent 1200						
Column	Luna-C18(2) 2.0*50mm, 5µm						
Mobile phase A (MPA)	H ₂ O+0.037 % (v/v) TFA						
Mobile phase B (MPB)	ACN+0.018 % (v/v) TFA						
Flow rate	0.8 mL/min						
Gradient Ratio	Time (min)	0.00	0.40	3.40	3.85	3.86	4.50
	MPA (%)	99	99	10	0	99	99
	MPB (%)	1	1	90	100	1	1
Detection	220 nm						