2-Aroylindoles and 2-Aroylbenzofurans with *N*-Hydroxyacrylamide Substructures as a Novel Series of Rationally Designed Histone Deacetylase Inhibitors†

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Histone deacetylase (HDAC) inhibitors are considered to be drugs for targeted cancer therapy and second-generation HDIs are currently being tested in clinical trials. Here, we report on the synthesis and biological evaluation of a novel HDAC inhibitor scaffold with the hydroxamate Zn^{2+} complexing headgroup, selected from the 2-aroylindol motif. Inhibition of nuclear extract HDAC and recombinant HDAC 1 as well as induction of histone $H3K^{9+14}$ hyperacetylation mediated by *E*-*N*-hydroxy-(2-aroylindole)acrylamides or *E*-*N*-hydroxy-(2-aroylbenzofuran)acrylamides were studied. Moreover, the cytotoxic activity, the effects on the cell cycle, and histone $H3S^{10}$ phosphorylation of selected compounds were determined. By use of a panel of 24 different human tumor cell lines, mean IC_{50} values of the most potent analogues **6c** and **7b** were 0.75 and 0.65 μM, respectively. The novel compounds were shown to be no substrates of the P-glycoprotein drug transporter. Comparable to N^1-hydroxy-N^8-phenyloctanediamide "**2** (SAHA)", cells in the S phase of the cell cycle are depleted, with partial arrest in G1 and G2/M and finally induction of massive apoptosis.

Introduction

Within the past decade our understanding of malignant cell growth and the underlying molecular alterations has offered new opportunities for molecular targeted cancer therapy. These new targeted approaches complement traditional chemotherapeutic drugs with the promise of a broader therapeutic window and efficacy in late-stage or therapy-resistant cancer patients. Carcinogenesis and tumor progression are controlled by both genetic and epigenetic events. Unlike genetic events, epigenetic aberrations such as DNA methylation evident in cancer cells can usually be reversed, thus reactivating epigenetically silenced genes.[1] On the basis of this rationale, the DNA methyltransferase (DNMT[a]) inhibitors 5-aza-3′-deoxycytidine **1a**[2] and 5-azacytidine **1b**,[2] acting as suicide inhibitors of DNMT1 and DNMT3a/b, are approved drugs for the treatment of patients with myelodysplastic syndrome[2] (MDS).

Another important epigenetic modification is based on the reversible acetylation of the N-terminal tails of core histone proteins H2A/B, H3, and H4.[3] In general, histone hyperacetylation is correlated with gene activation, whereas histone hypoacetylation mediates transcriptional repression.[4] The reversible acetylation and deacetylation of lysine residues are catalyzed by histone acetyltransferases (HATs) and histone deacetylases (HDACs), respectively. Mammalian HDAC isoenzymes fall into four distinct classes with at least 18 members.[5] HDAC class I (HDACs 1−3, 8), class II (HDACs 4−7, 9, 10), and class IV (HDAC11) enzymes are functionally related Zn^{2+}-containing amidohydrolases and are inhibited by the natural compound trichostatin A (TSA). They are part of multiprotein complexes examplified by interaction with transcriptional corepressors and transcription factors.[6] The function of HDAC class I enzymes in normal and pathological conditions was intensively studied. For example, targeted deletion of HDAC1 is followed by embryonic lethality in mice,[7] and HDAC 1−3 isoenzymes are highly expressed in prostate adenocarcinoma with HDAC 2 expression as an independent prognostic factor correlating with survival.[8] HDAC class II isoenzmyes are less well characterized, with HDAC 6 having substrates different from histone proteins like the mitotic spindle protein α-tubulin.[9] HDAC inhibitors (HDIs) effect the transcriptional regulation and induce or repress genes involved in differentiation, proliferation, cell cycle regulation, protein turnover, and apoptosis.[10] As such, they are considered as drugs for targeted cancer therapy.[11] HDAC inhibiting compounds are highly divergent in structure and comprise short-chain fatty acids and derivatives, hydroxamic acids, cyclic tetrapeptides/peptolides, benzamides, and others. Various agents are currently in clinical development, namely, the hydroxamte analogues (*E*)-3-(4-(((2-(1*H*-indol-3-yl)ethyl)-(2-hydroxyethyl)amino)methyl)phenyl)-*N*-hydroxyacrylamide "**1c** (LBH589)" (phase I, Novartis[12]), *N*-hydroxy-3-(3-(*N*-phenylsulfamoyl)phenyl)acrylamide "**1d** (PXD101)" (phase II, Topotarget/Curagen[13]), N^1-hydroxy-N^8-phenyloctanediamide "**2** (SAHA, vorinostat (Zolinza))" (phase II/III, Merck Inc.),[14] and *N*-(2-(4-(hydroxycarbamoyl)phenoxy)ethyl)-3-(isopropylamino)benzofuran-2-carboxamide hydrochloride "**3** (CRA024781)" (phase I, Pharmacyclics Inc.[15]), the benzamide analogues (4-(2-aminophenylcarbamoyl)benzylamino)methyl nicotinate "**3a** (MS275)" (phase II, Bayer-Schering AG[16]) and *N*-(2-aminophenyl)-4-((4-(pyridin-3-yl)pyrimidin-2-ylamino)methyl)benzamide "**3b** (MGCD0103)" (phase II, Methylgene[17]), the cyclic peptolide "**3c** (depsipeptide/

† This paper is dedicated to Professor Wolfgang Wiegrebe on the occasion of his 75th birthday.

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[a] Abbreviations:: ARK-B, Aurora kinase B; BOP, (benzotriazol-1-yloxy)-tris(dimethylamino)phosphonium hexafluorophosphate; CTCL, cutaneous T-cell lymphoma; DNMT, DNA methyltransferase; HAT, histone acetyltransferases; HDAC, histone deacetylases; HDI, histone deacetylase inhibitor; MDS, myelodysplastic syndrome; NFkB, nuclear factor κ B; NH_2OTHP, *O*-(tetrahydropyran-2-yl)hydroxylamine; NSCLC, non-small-cell lung cancer; PBS, phosphate buffered saline; STAT, signal transducer and activator of transcription; Pgp, P-glycoprotein; SAHA, suberoylanilidhydroxamic acid; SDS−PAGE, sodium dodecyl sulfate polyacrylamide gel electrophoresis; TSA, trichostatin A.

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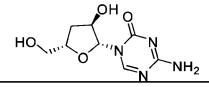
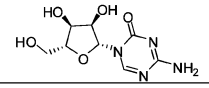
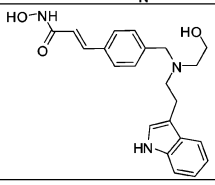
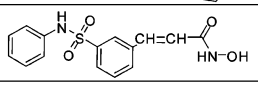
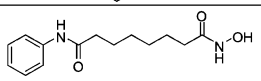
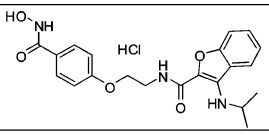
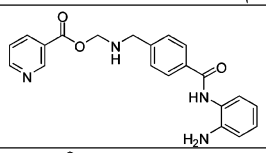
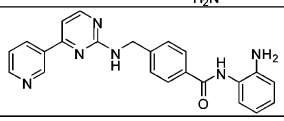
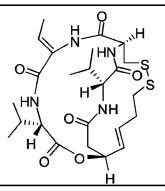
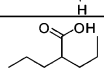
Table 1. Chemical Structures and Chemical Classes, Their Mode-of-Action, Specificity, and Development Status

Formula	Nr.	Name and chemical class	Mode-of-action	Specificity	Status
	**1a**	Decitabine, deoxycytidine analog	DNMT suicide inhibitor	DNMT-1, DNMT-3a/b	approved drug for MDS therapy
	**1b**	Vidaza, azacytidine analog	DNMT suicide inhibitor	DNMT-1, DNMT-3a/b	approved drug for MDS therapy
	**1c**	LBH589 hydroxamate analog	Substrate competitive HDI	pan HDI (class I, II and IV)	Clinical phase 1
	**1d**	PXD101, hydroxamate analog	Substrate competitive HDI	pan HDI (class I, II and IV)	Clinical phase 1 / 2
	**2**	SAHA, hydroxamate analog	Substrate competitive HDI	pan HDI (class I, II and IV)	Approved drug for CTCL therapy
	**3**	CRA024781. hydroxamate analog	Substrate competitive HDI	pan HDI (class I,II and IV)	IND filed
	**3a**	MS275, benzamide analog	Substrate competitive HDI	Isotype selective (class I)	Clinical phase 1 / 2
	**3b**	MGCD0103, benzamide analog	Substrate competitive HDI	Isotype selective (class I)	Clinical trials phase 1 / 2
	**3c**	Depsipeptide / FK228, peptolid (natural compound from *chromobacterium violaceum*)	Pro-drug, reduced thiol analog acts as substrate competitive HDI	Isotype selective (class I)	Approval for CTCL therapy ongoing
	**3d**	Valproic acid / VPA, butyrate analog	Weak substrate competitive HDI		Clinical trials phase 1 / 2; approved for CNS disorders

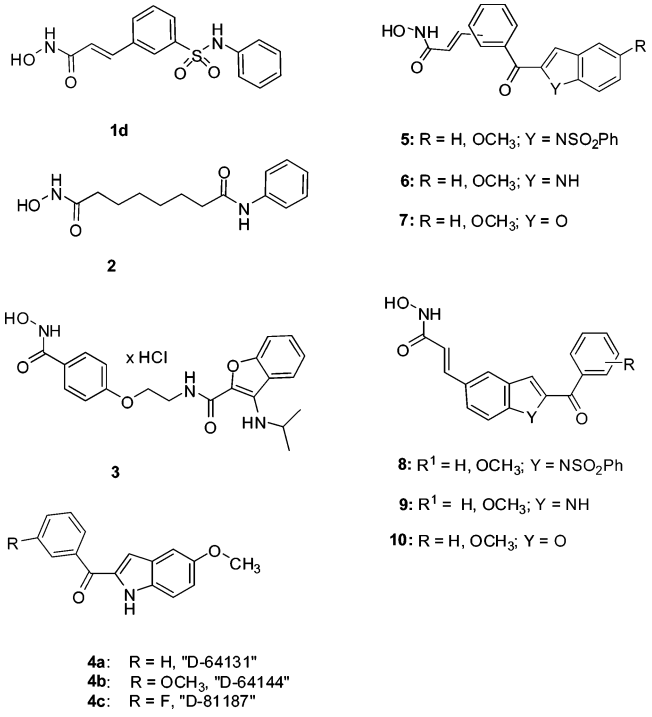
FK228)" (phase II, Gloucester/NCI[18]), and finally butyrate analogues like valproic acid "**3d** (VPA)" (phase II, Topotarget[19]). Suberoylanilidhydroxamic acid (**2**) was recently approved for treatment of refractory cutaneous T-cell lymphoma (CTCL[20]). It is important to note that except for benzamide analogues **3a** and **3b**, all hydroxamate-based HDIs in clinical development are unselective inhibitors of HDAC class I and class II enzymes. Nevertheless, it appears to be a common property of unselective as well as class I selective HDIs to induce alterations of gene expression with a similar final outcome, namely, cell cycle alterations, induction of differentiation, and apoptosis.[1,21] In contrast to antimitotic agents, proliferating as well as arrested (dormant) tumor cells are killed with similar potency.[21,22] From recent studies it is now evident that the antitumor activity of HDIs cannot be explained by epigenetic mechanisms alone. In melanoma cells, interaction of NFκB p65 with STAT1 (signal transducer and activator of transcription) is dependent on CBP (CRE binding protein)/HDAC regulated STAT1 acetylation controlling the nuclear translocation and antiapoptotic function of NFκB.[23] In another study, interaction of HDAC 3 with the mitotic kinase Aurora B (ARK-B) and HDAC 3 dependent phosphorylation of histone H3S[10] by ARK-B were shown, giving a hint to the partial M-phase arrest seen by many HDIs.[24] An overview of the reference compounds as discussed with their mode-of-action and specificity is compiled in Table 1.

Here, we report on a novel HDAC inhibitor scaffold with the hydroxamate Zn^{2+} complexing headgroup ("warhead"). The new scaffold was selected from the 2-aroylindol motif (Chart 1), described to be highly active as an antimitotic agent by destabilizing tubulin polymerization.[25] Combining both structural elements abolished the tubulin binding activity, leading to HDAC inhibitors with an in vitro potency equal or superior to **2** as the most advanced HDAC inhibitor.

Chemistry

The synthetic route to obtain the desired target compounds is given in the following: The substituted 2-aroylindole derivatives **13a**−**c** were easily prepared by coupling the lithiated indole

Chart 1. Chemical Structures of Selected Hydroxamate-Based HDAC Inhibitors in Clinical Development (**1**−**3**), Selected Aroylindoles (**4**), and Novel Rationally Designed Compounds (**5**−**10**)



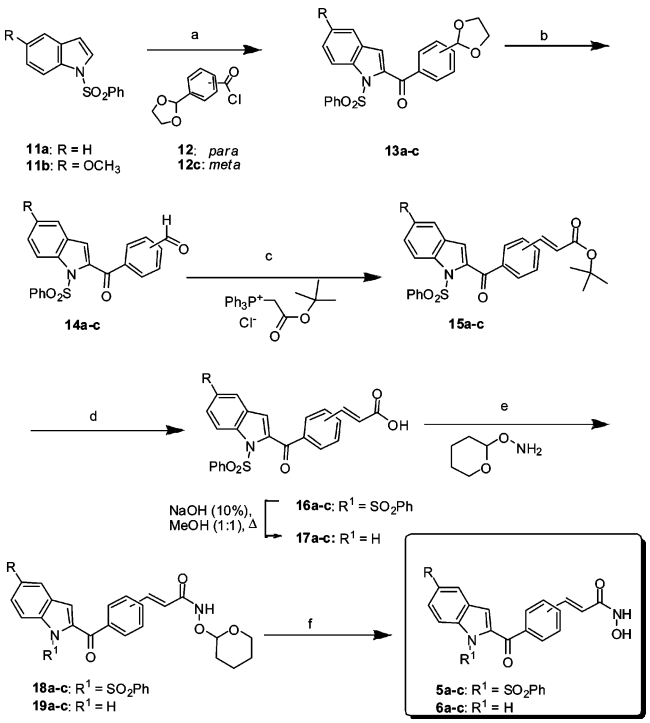
species with the respective acid chlorides (**12, 12c**) bearing a protected aldehyde group. Deprotection of the aldehydes (**13a**−**c**) with perchloric acid followed by Wittig olefination[26] and cleavage of the resulting trans-oriented acrylic acid *tert*-butyl esters (**15a**−**c**) with trifluoroacetic acid yielded the corresponding acrylic acids (**16a**−**c**). To obtain the N-dephenylsulfonated carboxylic acids (**17a**−**c**), deprotection with NaOH in water/methanol was performed. Amidation of **16a**−**c** or **17a**−**c** with commercially available NH_2OTHP (*O*-(tetrahydropyran-2-yl)-hydroxylamine) by use of BOP {(benzotriazol-1-yloxy)tris-(dimethylamino)phosphonium hexafluorophosphate} as a coupling reagent[27] and cleavage of the tetrahydropyran-2-yl protected acrylamides (**18a**−**c** and **19a**−**c)** led to the *N*-hydroxyacrylamides **5a**−**c** and **6a**−**c**.

The respective acid chlorides **12** and **12c**, which were used for the coupling reaction described in Scheme 1, were prepared as shown in Scheme 2 by protection of the commercially available formylbenzoic acids (**20**, **20c**) with ethylene glycol according to Kamitori[28] and by treatment of the resulting [1,3]-dioxolan-2-ylbenzoic acids (**21**, **21c**) with thionyl chloride in THF/pyridine solution.

Following the strategy described in Scheme 1, the *N*-hydroxyacrylamide substituent was introduced at C-5 of the indole nucleus (Scheme 3). Lithiation of 1-benzenesulfonyl-5-[1,3]dioxolan-2-yl-1*H*-indole[29] (**24**), coupling with the respective acid chlorides, and cleavage of the resulting [1,3]dioxolanes (**25a**,**b**) by perchloric acid yielded the aldehydes (**26a**,**b**), which were converted to the *E*-*N*-hydroxyacrylamides (**8a**,**b** and **9a**,**b**) in four steps.

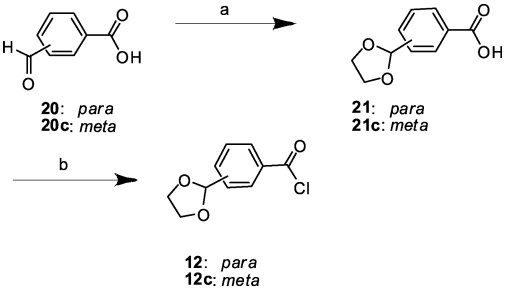
The corresponding benzofuran-2-carbonylbenzaldehydes (**34a**,**b**) were prepared from salicylic aldehyde (**32**) and the respective substituted ω-bromoacetophenones (**33a**,**b**) under basic conditions in acetonitrile (Scheme 4) according to Stille et al.[30] The ω-bromoacetophenones (**33a**,**b**) thereby were prepared by bromination in CH_2Cl_2 with bromine. Introduction of the

Scheme 1. Synthesis of *E*-*N*-Hydroxy-(2-aroylindole)acrylamides[a]



[a] (a) BuLi, −78 °C, THF; (b) $HClO_4$ (37%), CH_2Cl_2, 0 °C, 2 h; (c) NaOH, NEt_3, H_2O, 20 °C; (d) (1) CF_3COOH, 20 °C, (2) H_2O; (e) BOP, DMF, NEt_3, room temp, 1 h; (f) HCl, MeOH, 20 °C, 24 h.

Scheme 2. Synthesis of Acid Chlorides (**12**, **12c**) Bearing a Protected Aldehyde Group[a]



[a] (a) 1,2-ethandiol, Al_2O_3, toluene, Δ, 48 h; (b) pyridine, $SOCl_2$, 20 °C, 30 min.

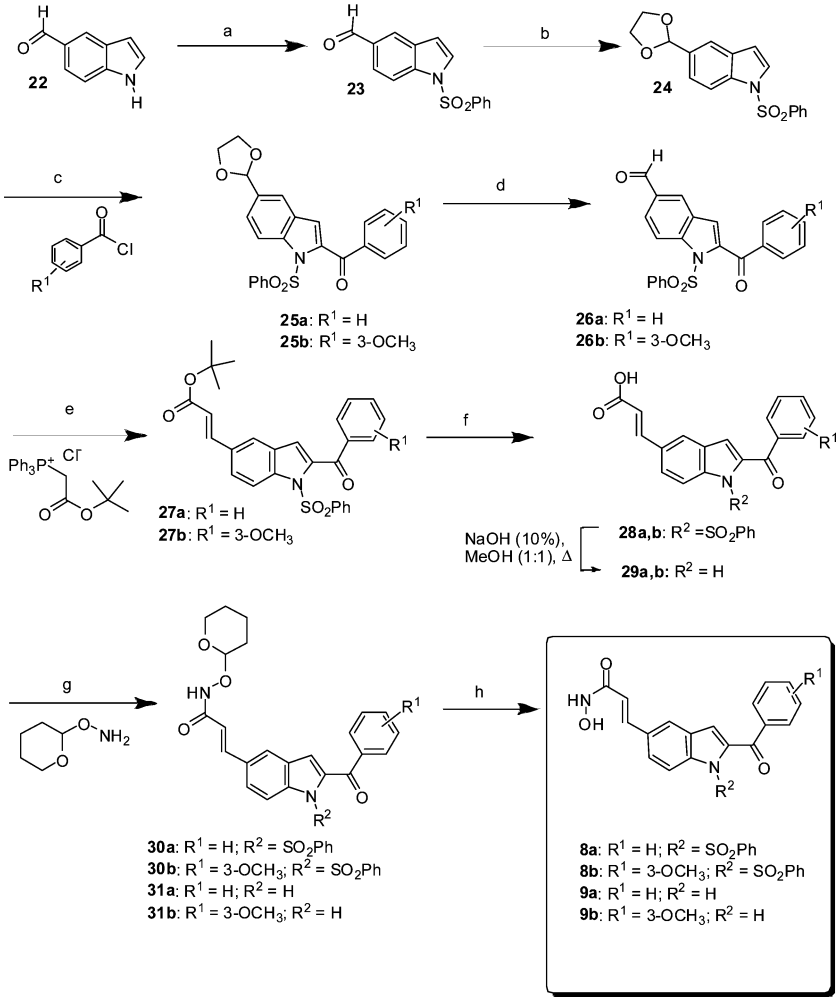
N-hydroxyacrylamide substituent was performed in the same manner (Scheme 4) as described for the indole nucleus and led to the desired compounds **36a** and **36b**.

Again, introduction of the *N*-hydroxyacrylamide substituent was performed at C-5 of the 2-aroylbenzofuranyl nucleus (Scheme 5) following the general synthetic strategy described in Scheme 4. Thus, 4-hydroxybenzene-1,3-dicarbaldehyde (**38**) used in this sequence was easily accessible from 4-hydroxybenzaldehyde (**39**) according to Stille.[30]

Results and Discussion

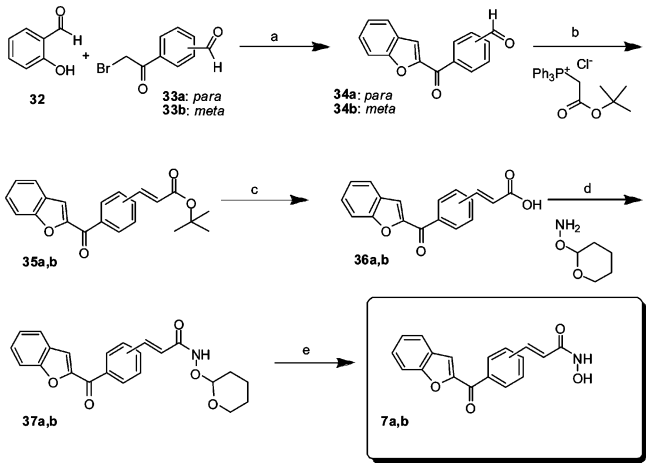
The in vitro data for inhibition of HeLa nuclear extract HDAC activity (a mixture of HDAC isoenzymes 1−3, 5, and 8) and recombinant HDAC 1 (rHDAC 1) in comparison with **2** as a reference compound are compiled in Tables 2−5. To assess the cellular efficacy of HDAC inhibition, EC_{50} and IC_{50} values for the induction of histone H3K(Ac)$^{9+14}$ hyperacetylation and cytotoxicity vs HeLa cells are also given. Mean data from two experiments are shown. Respective concentration−effect curves derived from the histone H3 hyperacetylation assay for selected

Scheme 3. Preparation of *E*-3-(2-Benzoyl-1*H*-indol-5-yl)-*N*-hydroxyacrylamides[a]



[a] (a) NaOH, CH_2Cl_2, $PhSO_2Cl$; (b) 1,2 ethandiol, Al_2O_3, toluene, Δ, 48 h; (c) *n*-BuLi, −78 °C, THF; (d) $HClO_4$ (37%), CH_2Cl_2, 0 °C, 2 h; (e) NaOH, NEt_3, H_2O, 20 °C; (f) (1) CF_3COOH, 20 °C; (2) H_2O; (g) BOP, DMF, NEt_3, room temp, 1 h; (h) HCl, MeOH, 20 °C, 24 h.

Scheme 4[a]



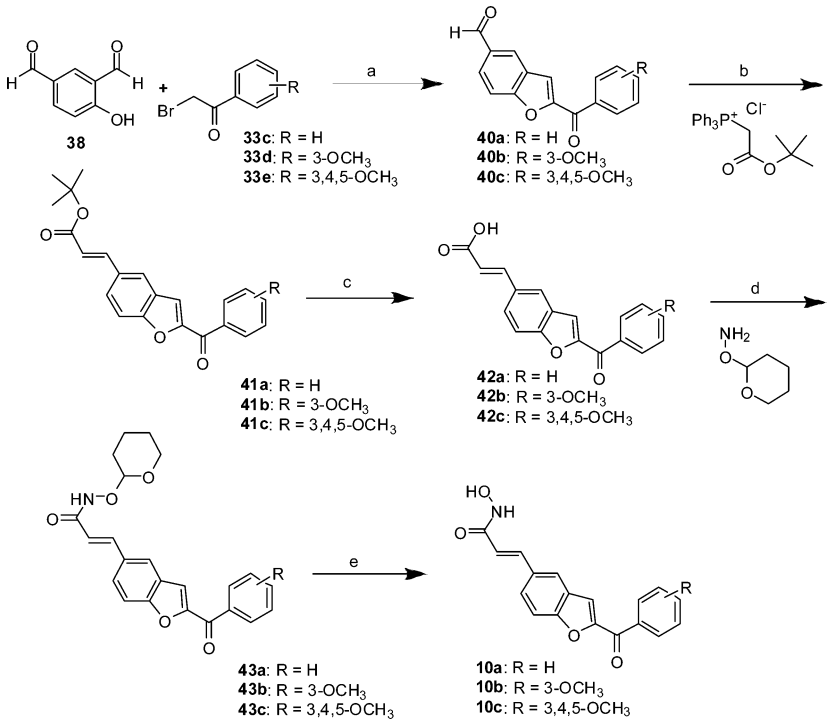
[a] (a) K_2CO_3, CH_3CN, Δ; (b) NaOH, NEt_3, H_2O, 20 °C; (c) (1) CF_3COOH, 20 °C, (2) H_2O; (d) BOP, DMF, NEt_3, room temp, 1 h; (e) HCl, MeOH, 20 °C, 24 h.

compounds (**2**, **6c**, **7b**, **9a**, and **10c**) are shown in Figure 1. Combining the structural features of 2-aroylindoles with an *E*-*N*-hydroxyacrylamide motif leads to compounds with a concentration-dependent inhibition of nuclear extract HDAC and rHDAC 1 in the submicromolar range and significant H3K hyperacetylation correlating well with cytotoxicity against HeLa cells.

When we investigated 2-aroylindoles **4a**−**c**,[25] removal of the *N*-phenylsulfonyl protection group resulted in 40- to 100-fold higher activity mediated by inhibition of tubulin polymerization. Here, however, no effect of the phenylsulfonyl group on biological activity can be observed when comparing derivatives **5a**−**c** with their corresponding N-unsubstituted derivatives **6a**−**c**. Interestingly, by comparison of the biological data for the compounds being substituted by the trans-oriented *E*-*N*-hydroxyacrylamide motif in position 4 (**5a** and **6a**) with the respective compounds being substituted in position 3 of the benzoyl system (**5c** and **6c**), the change in position led to about 10-fold higher inhibition of nuclear extract HDAC and rHDAC 1 activity as well as to a significant increase of H3K hyperacetylation and cytotoxicity. When the biological data for the unsubstituted compounds **5a** and **6a** were compared with data for **5b** and **6b**, substitution in position 5 of the indolyl system by a methoxy group (**5b** and **6b**), again in contrast to the findings for the 2-aroylindole compounds **4a**−**c**,[25] did not exhibit any effects. When these effects and the results obtained for the *E*-*N*-hydroxy-(2-aroylbenzofuran)acrylamides **7a** and **7b** (Table 3) were evaluated, the same trend is seen. For the carboxylic acids (**16c**, **17c**) structurally related to the most potent hydroxamates (**5c**, **6c**), no significant inhibition of HDAC has been observed.

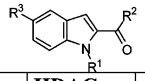
Compounds **6c** and **7b** were also tested for inhibition of rHDAC 3 and 8 isoenzymes as additional class I representatives. rHDAC 3 and 8 were inhibited by **6c** and **7b** with IC_{50} values

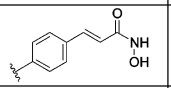
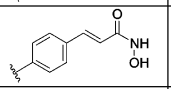
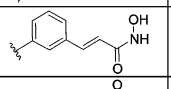
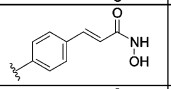
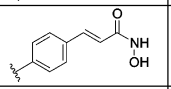
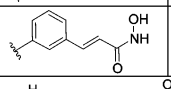
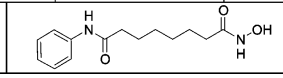
Scheme 5. Preparation of *E*-3-(2-Benzoylbenzofuran-5-yl)-*N*-hydroxyacrylamides **10a**−**c**[a]



[a] (a) K_2CO_3, CH_3CN, Δ; (b) NaOH, NEt_3, H_2O, 20 °C; (c) (1) CF_3COOH, 20 °C, (2) H_2O; (d) BOP, DMF, NEt_3, room temp, 1 h; (e) HCl, MeOH, 20 °C, 24 h.

Table 2. Inhibition of Nuclear Extract HDAC and rHDAC 1 and Induction of H3K(Ac)$^{9+14}$ Hyperacetylation and Cytotoxicity Against HeLa Cells Mediated by *E*-*N*-Hydroxy-(2-aroylindole)acrylamides[a]



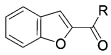
ID	R^1	R^2	R^3	HDAC inhibition IC_{50} [μM]	rHDAC 1 inhibition IC_{50} [μM]	H3K(Ac)$^{9+14}$ induction EC_{50} [μM]	Cytotox HeLa IC_{50} [μM]
5a	SO_2Ph		H	0.27	0.69	5.70	5.42
5b	SO_2Ph		OCH_3	0.26	0.88	6.90	5.42
5c	SO_2Ph		H	0.034	0.089	1.00	1.73
6a	H		H	0.31	0.41	2.97	4.75
6b	H		OCH_3	0.28	0.32	4.27	3.44
6c	H		H	0.039	0.031	1.30	0.85
2				0.070	0.020	6.67	1.75

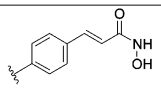
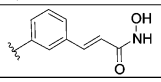
[a] Mean data from at least two experiments are shown.

of 0.05 μM/0.032 μM and 0.079 μM/0.079 μM, respectively. Thus, both compounds behave as pan HDAC inhibitors like **2**.

Substitution by the *N*-hydroxyacrylamide motif in position 5 of the indolyl sytem (Tables 4 and 5, compounds **8a**−**10c**) also led to potent HDAC inhibition and cellular activity. Inhibition of HDAC enzymatic activity was the most distinct in the case of the N-phenylsulfonated indolyl compounds **8a**,**b**, followed by **9a**,**b** and the benzofuranyl compounds **10a**−**c**. Reflecting the potency in inhibition of nuclear extract HDAC and rHDAC 1, cytotoxicity against HeLa cells is highest for **8a** and **8b**. Substitution patterns in the benzoyl system had no significant effect on biological activity, as seen for **8a** and **8b** or **10a** and **10b**. Considering the fact that small-molecule HDAC inhibitors in general are known to be built from a cap group, a spacer, and a Zn^{2+} chelating headgroup like the hydroxamic acid motif, this can be most likely explained by an outside orientation of

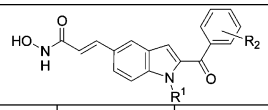
Table 3. Inhibition of Nuclear Extract HDAC and rHDAC 1 and Induction of H3K(Ac)$^{9+14}$ Hyperacetylation and Cytotoxicity Against HeLa Cells of *E*-*N*-Hydroxy-(2-aroylbenzofurane)acrylamides **7a** and **7b**[a]



ID	R	HDAC inhibition IC_{50} [μM]	rHDAC 1 inhibition IC_{50} [μM]	H3K(Ac)$^{9+14}$ induction EC_{50} [μM]	Cytotox HeLa IC_{50} [μM]
7a		0.45	0.52	1.10	2.60
7b		0.057	0.052	0.29	0.68

[a] Mean data from at least two experiments are shown.

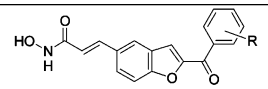
Table 4. Inhibition of Nuclear Extract HDAc and rHDAC 1 and Induction of H3K(Ac)$^{9+14}$ Hyperacetylation and Cytotoxicity Against HeLa Cells of *E*-3-(2-Benzoyl-1*H*-indol-5-yl)-*N*-hydroxyacrylamides **8a**,**b** and **9a**,**b**[a]



ID	R^1	R^2	HDAC inhibition IC_{50} [μM]	rHDAC 1 inhibition IC_{50} [μM]	H3K(Ac)$^{9+14}$ induction EC_{50} [μM]	Cytotox HeLa IC_{50} [μM]
8a	SO_2Ph	H	0.09	0.20	2.67	2.88
8b	SO_2Ph	3-OCH_3	0.12	0.23	2.78	3.34
9a	H	H	0.34	0.25	3.77	3.97
9b	H	3-OCH_3	0.33	0.30	2.57	2.96

[a] Mean data from at least two experiments are shown.

Table 5. Inhibition of Nuclear Extract HDAC and rHDAC 1 and Induction of H3K(Ac)$^{9+14}$ Hyperacetylation and Cytotoxicity on HeLa Cells of *E*-3-(2-Benzoylbenzofuran-5-yl)-*N*-hydroxyacrylamides **10a**−**c**[a]



ID	R	HDAC inhibition IC_{50} [μM]	rHDAC 1 inhibition IC_{50} [μM]	H3K(Ac)$^{9+14}$ induction EC_{50} [μM]	Cytotox HeLa IC_{50} [μM
10a	H	0.83	1.20	2.65	5.61
10b	3-OCH_3	0.65	0.95	4.10	4.11
10c	3,4,5-OCH_3	1.0	1.10	7.50	5.77

[a] Mean data from at least two experiments are shown.

the benzoyl system as the cap group. As a consequence, the cap group interacts with the solvent accessible surface of the HDAC enzyme, which is in proximity to the active site as shown for hydroxamate analogues, binding to the class I isoenzyme HDAC 8.[31] Since this region of the HDAC enzyme is highly malleable, modification of the aromatic system would result only in a minor influence on the binding affinity.

The biological profile of the most potent analogues **6c** and **7b** with the indolyl or benzofuran scaffold was investigated in more detail. The antiproliferative/cytotoxic activity was determined by using a broad panel of 24 different human tumor cell lines emenating from 16 different solid and hematological tumor entities (Table 6). The mean IC_{50} values were calculated as 0.75 μM for **6c** and 0.65 μM for **7b**, which is about 5 times more potent as **2** with a mean IC_{50} of 3.3 μM. Cell lines with the highest sensitivity (IC_{50} < 0.3 μM) are K562 (chronic myeloid leukemia/CML), EOL1 (acute myeloid eosinophilic leukemia/AML), CCRF-CEM (acute T-lymphoblastic leukemia/ALL), and PC3 (androgen-independent prostate carcinoma). Both analogues are no substrate of the P-glycoprotein (P-gp) drug transporter, since the sensitivities toward vincristine-resistent P-gp overex-

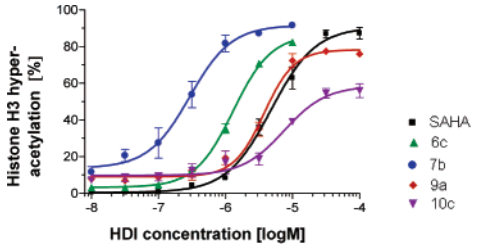
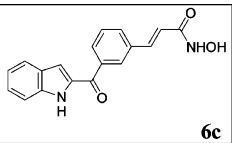
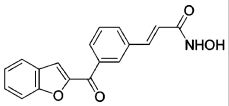


Figure 1. Induction of histone H3K hyperacetylation in HeLa cells. HeLa cervical carcinoma cells were treated with compounds **6c** (▲), **7b** (●), **9a** (◆), **10c** (▼), and SAHA/**2** (■) as reference for 24 h before fixation, staining with a H3K(Ac)$^{9+14}$ specific antibody and using single cell image analysis with the Cellomics array scan device. EC_{50} values were calculated by nonlinear regression analysis using the program GraphPad prism.

Table 6. Cytotoxicity Profile[a]

	 **6c**	 **7b**	**2**
Cell line	**IC_{50} [μM]**	**IC_{50} [μM]**	**IC_{50} [μM]**
NCI-H460	0.26	0.25	1.2
A549	0.97	0.77	1.8
NCI-H69	0.33	0.19	nd
MCF7	1.9	1.3	6.7
MDA-MB-468	0.55	0.50	1.3
MDA-MB-231	0.9	0.73	2.2
SK-BR-3	0.53	0.31	2.6
SK-OV-3	0.90	0.74	2.2
A-2780	0.34	0.35	nd
RKO p21 (not proliferating)	2.57	3.69	3.81
RKO p21 (proliferating)	1.05	0.96	1.76
RKO p21 (40% human serum)	4.9	5.0	nd
HCT-15	0.77	0.74	3.9
HCT-116	0.36	0.36	nd
PC-3	0.21	0.16	1.9
AsPC-1	3.9	3.50	≈ 100
HeLa	0.65	0.49	1.5
Cal 27	0.75	0.66	3.2
A-431	0.60	0.59	4.2
Hec-1-A	0.62	0.47	1.0
Saos-2	1.0	0.92	2.3
U-87 MG	1.0	1.1	11.0
WM 266-4	0.31	0.31	nd
K-562	0.17	0.15	0.67
EOL1	0.073	0.049	1.0
CCRF-CEM	0.20	0.18	0.80
CCRF-CEM (Vincristine resistant)	0.25	0.24	0.95
Mean IC_{50}	**0.75**	**0.65**	**3.3**

[a] The antiproliferative, cytotoxic activity of selected compounds was evaluated using a panel of 24 different human tumor cell lines. Further information on the different cell lines is given in the Experimental Section. nd: not determined.

pressing CCRF-CEM cells and the parental cell line are similar. Cytotoxicity toward RKO colon carcinoma cells is only slightly affected by addition of 40 vol % human serum, pointing toward low serum protein binding. Finally, **6c**, **7b**, and **2** depict a cell cycle independent cytotoxic activity as shown by using the RKOp21^{waf1} model.[32] Proliferation of arrested p21^{waf1} expressing and proliferating RKO cells is inhibited with nearly equal potency (Figure 2).

The effect of **6c** and **7b** on the cell cycle of A549 non-small-cell lung cancer cells (NSCLC) by flow cytometry was studied next. Asynchronous proliferating A549 cells were treated for 24 h before staining of nuclear DNA with propidium iodide. As shown in Figure 3, both compounds at ≥1 μM deplete cells in the S-phase (>2 N and <4 N DNA content). A partial arrest and transient in G1 (2 N DNA content) and G2/M (4 N DNA

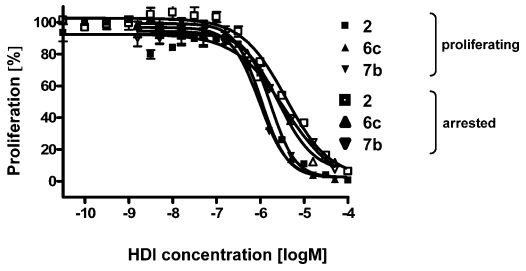


Figure 2. Cytotoxicity in the RKOp21^{waf1} cell line model. The effect of **2** (□), **6c** (△), and **7b** (▽) on arrested and proliferating RKOp21^{waf1} cells after treatment for 72 h was determined using the Alamar blue assay. IC_{50} values were calculated by nonlinear regression analysis using the program GraphPad prism.

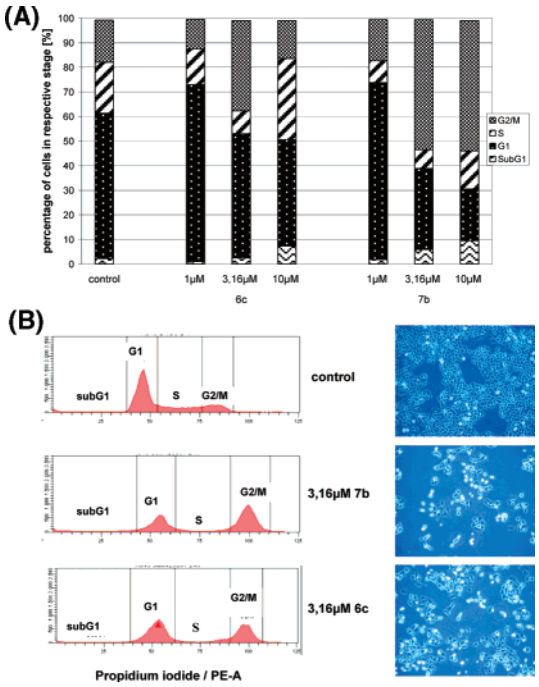


Figure 3. Flow cytometric analysis of A549 NSCLC cells. A549 cells treated for 24 h with **6c** and **7b** at selected concentrations were analyzed for cell populations in the G1, S, and G2/M stages of the cell division cycle. Also, apoptotic cells with a subG1 DNA content were determined. A column representation is given in part A, and selected histograms and cell pictures by phase contrast microscopy are shown in part B.

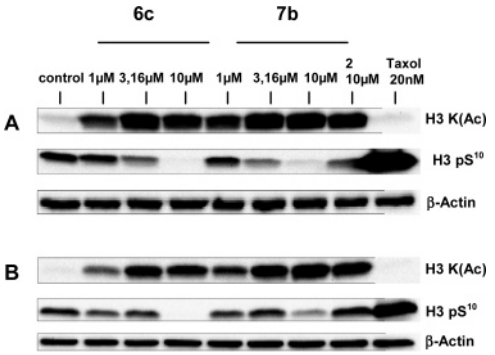


Figure 4. Analysis of histone $H3K^{9+14}$ acetylation and $H3S^{10}$ phosphorylation in RKO and A549 cells. RKO colon carcinoma (A) and A549 NSCLC (B) cell lines were treated with selected concentrations of **6c** and **7b** for 16 h before cell lysis and Western blot analysis, detecting histone $H3K(Ac)^{9+14}$ acetylation and $H3S^{10}$ phosphorylation. Samples of 10 μM SAHA/**2** and 20 nM paclitaxel as an tubulin stabilizing, antimitotic agent were included as controls.

content) are evident at 3.2 μM (Figure 3B), with massive apoptosis (subG1 cell population, <2 N DNA content) at 10 μM. This massive apoptosis affected the cell cycle analysis by an overestimation of cells in the S and G1 phases (Figure 3A).

SAHA (**2**) as a reference behaved quite similarly, also depleting S phase cells and partially arresting at G1 and G2/M (data not shown). Finally, histone $H3S^{10}$ phosphorylation as a marker for ARK-B activation and mitotic arrest was studied in RKO and A549 cells. As shown in Figure 4, **6c** and **7b** induce a strong histone H3 hyperacetylation with maximum levels reached in both cell lines between 1 and 3 μM. In parallel, histone $H3S^{10}$ phosphorylation was nearly completely inhibited at 10 μM and below the level of untreated, cycling control cells. The opposite is true for the antimitotic agent paclitaxel, showing a strong induction of histone $H3S^{10}$ phosphorylation as a hallmark of mitotic arrest. Since both HDIs induce a partial arrest in G2/M, this result is unexpected. One possible explanation is an arrest in G2. Another hypothesis is based on the concept that only hypoacetylated histone H3 N-terminal tails are good substrates for ARK-B as described recently by Li et al.[24] Since **6c** and **7b** potently induce H3 hyperacetylation, this antagonizes ARK-B activity and as a consequence causes low or undetectable histone $H3S^{10}$ phosphorylation.

Conclusion

We conclude that 2-aroylindoles as **6c** and 2-aroylbenzofurans as **7b** with the hydroxamate headgroup constitute a new class of potent HDAC class I/II inhibitors with a cell-cycle-independent and broad cytotoxicity superior to that of SAHA (**2**) as an approved drug for cutaneous T-cell lymphoma treatment.

Experimental Section

Biological Methods. Biochemical HDAC Assays. HDAC activity was determined by using nuclei isolated from the cervical carcinoma cell line HeLa (ATCC CCL2) according to a modified method first described by Dignam et al.[33] For recombinant HDAC1 expression, a clonal HEK293 (ATCC CRL1573) human kidney cell line expressing the human HDAC1 isoenzyme bearing a C-terminal FLAG epitope was provided by E. Verdin, The Gladstone Institute, San Francisco, CA. The rHDAC1 protein was purified by M2 affinity gel chromatography according to the manufacturer's protocol (Sigma no. A2220). Purified HDAC protein samples were routinely analyzed by SDS−PAGE (12.5% or 10% Laemmli gels) followed by Coomassie stain and Western blotting using a FLAG-specific antibody (anti-M2-POX antibody, Sigma no. A8592) followed by ECL detection (GE Healthcare). In addition, protein batches were analyzed by Western blotting for various HDAC isoenzymes including HDAC1.

The biochemical HDAC activity assay was essentially done as described by Wegener et al.[34] The overall reproducibility was very high (standard error of the mean in the used test systems was ≤25%, and mean values of the test compounds in general were within the standard error of the mean of the test system). For details, see Supporting Information.

Cellular Histone H3 Hyperacetylation Assay. To assess the cellular efficacy of a histone deacetylase inhibition, an assay was set up for use on the Cellomics "ArrayScan II" platform for a quantitative calculation of histone acetylation as described.[35] The overall reproducibility was very high (standard error of the mean in the used test systems was ≤25%, and mean values of the test compounds in general were within the standard error of mean of the test system). For details, see Supporting Information.

Cellular Proliferation Assay. The antiproliferative cytotoxic activity of selected compounds was evaluated using the tumor cell lines HeLa (cervical carcinoma, ATCC CCL-2), H460 (non-small-cell lung cancer, ATCC HTB-177), A549 (NSCLC, ATCC CCL-185), MCF7 (breast carcinoma, ATCC HTB-22), MDA-MB468 (breast carcinoma, ATCC HTB-132), MDA-MB435 (breast carcinoma, ATCC HTB-129), MDA-MB231 (breast carcinoma, ATCC HTB-26), SKBR-3 (breast carcinoma, ATCC HTB-30), SKOV-3 (ovarian carcinoma, ATCC HTB-77), RKOp21 (colon carcinoma[32]), HCT-15 (colon carcinoma, ATCC CCL-225), PC3 (prostate carcinoma, ATCC CRL-1435), AsPC1 (pancreatic carcinoma, ATCC CRL-1682), Cal27 (tongue carcinoma, ATCC CRL-2095), A-431 (vulva carcinoma, ATCC CRL-2592), Hec1A (endometrial carcinoma, ATCC HTB-112), Saos-2 (osteosarcoma, ATCC HTB-85), U87MG (glioblastoma, ATCC HTB-14), WM266−4 (melanoma, ECACC 91061233), K562 (chronic myeloid carcinoma, DSZM ACC 10), EOL1 (acute hypereosinophilic myeloid leukemia, DSZM ACC386), and CCRF-CEM and CCRF-CEM VCR1000 (acute lymphoblastic leukemia sensitive and resistant toward vincristine, DSZM 240). For quantification of cellular proliferation/cytotoxicity, the Alamar blue (resazurin) cell viability assay was applied.[36] Cell lines were seeded at their respective density into

96-well flat bottom plates (Costar no. 3598) in a volume of 200 μL per well. Twenty-four hours after seeding, 1 μL each of the compound dilutions was added in duplicate and incubation continued for 72 h at 37 °C in a humidified atmosphere containing 5% CO_2. Finally, an amount of 20 μL of resazurin solution (90 mg/mL in PBS) was added, and after 4 h of incubation at 37 °C, the fluorescence was measured at an excitation λ of 544 nm and an emission λ of 590 nm using a Wallac Victor V reader. For calculation of cell viability, the emission from untreated cells was set to 100% and from medium-only samples to 0% viability. The viability in % was calculated using the formula

$$\frac{E_{590\text{nm}}X - E_{590\text{nm}}C_0}{E_{590\text{nm}}C_{100} - E_{590\text{nm}}C_0} \times 100$$

Flow Cytometric Analysis. A549 NSCLC cells were seeded at 5×10^5 cells/10 cm cell culture dish 24 h before treatment to allow logarithmic proliferation during the experiment. After 24 h, cells were treated with test compound (after an additional 24 and 48 h), the culture medium was collected, and floating cells were collected by short centrifugation (5 min at 1200 rpm). Attached cells were washed once with 5 mL of PBS, detached by treatment with 3 mL of trypsin/EDTA solution (short incubation for 3−5 min), and together with detached floating cells, transferred into 15 mL Falcon tubes. The cell culture dish was washed with 10 mL of DMEM to collect all remaining cells. After centrifugation for 5 min at 1200rpm at room temperature, the supernatant was discarded and the cell pellet washed twice with 10 mL of PBS. The supernatant was discarded, the cells resuspended in 4 mL of ice-cold fixation solution (70 vol % ethanol, 30 vol% H_2O). Next an amount of 10 mL of PBS (4 °C) was added to the cells before centrifugation for 5 min at 1200rpm. The supernatant was discarded, the cell pellet washed once with 10 mL of PBS (4 °C), and the cell pellet finally resuspended in 1 mL of staining solution (10 μg/mL RNAse A, 10 μg/mL propidiumiodide in PBS). After incubation for 30 min at room temperature in the dark, cells were stored on ice and analyzed using the Becton Dickinson FACS Canto device. Analysis was done at an exitation λ of 488 nm with emission set at a λ of 600 nm.

Western Blot Analysis. For Western blot analysis, 2×10^5/well A549 or 3×10^5/well RKOp21 carcinoma cells in six-well cell culture plates were treated with the test compounds for 16 h. Attached cells were lysed in 150 μL of lysis buffer (50 mM Tris-HCl, pH 8, 150 mM NaCl, 1 v/v NP-40, 0.5% w/v Na desoxycholate, 0.2% w/v disodium dodecyl sulfate (SDS), 0.02% w/v NaN_3, 1 mM Na vanadate, 20 mM NaF, 100 μg/mL PMSF, protease inhibitor mix/Roche, and 50 U/mL Benzonase) at 4 °C. Equal amounts of protein were separated by SDS−PAGE before transfer to polyvinylidene difluoride (PVDF) membrane (Biorad no. 162-0177) by semidry blotting.[37] The following antibodies were used: monoclonal antibody specific for β-actin (clone AC-12, Sigma no. A-5441), polyclonal antibody specific for $H3K(Ac)^{9+14}$ (Calbiochem no. 382158), polyclonal antibody specific for $H3S^{10}p$ (Cell Signaling no. 9701), goat antirabbit IgG-HRP conjugated (Biorad, 170-6515), and goat-anti-mouse IgG-HRP conjugated (Biorad, 170-6516).

Chemical Procedures. General. NMR spectra were recorded with a Bruker Avance 300 MHz spectrometer at 300 K, using TMS as an internal standard. IR spectra (KBr or pure solid) were measured with a Bruker Tensor 27 spectrometer. Melting points were determined with a Büchi B-545. MS spectra were measured with a Finnigan MAT 95 (EI, 70 eV, and HR-MS spectra) or with a Finnigan Thermo Quest TSQ 7000 (ESI) (CH_2Cl_2/MeOH + 10 mmol/L NH_4Ac). All reactions were carried out under nitrogen atmosphere. Elemental analyses were performed by the Analytical Laboratory of the University of Regensburg.

1-Benzenesulfonyl-1*H*-indole (11a) was prepared from indole (Acros) according to procedures earlier described.[38]

1-Benzenesulfonyl-5-methoxy-1*H*-indole (11b) was prepared from 5-methoxyindole (Biosynth) according to procedures[38] described earlier.

4-[1,3]Dioxolan-2-ylbenzoic acid (21) was prepared from 4-carboxybenzaldehyd (Acros) and ethane-1,2-diol according to the literature.[28]

1*H*-Indole-5-carbaldehyde (22)[39] was prepared from 5-bromoindole (Acros) according to the literature.[39]

1-Benzenesulfonyl-1*H*-indole-5-carbaldehyde (23) was prepared from 1*H*-Indole-5-carbaldehyde (**22**)[39] according to the literature.[29]

5-(1,3-Dioxolan-2-yl)-1-(phenylsulfonyl)-1*H*-indole (24) was prepared from **23** according to the analogous literature[28] as described for **21c**. ^{1}H NMR (DMSO-d_6): δ (ppm) = 8.25 (s, 1H), 8.17−8.16 (m, 2H), 8.10−7.93 (m, 4H), 7.78−7.51 (m, 3H), 4.11−4.09 (m, 2H), 4.08−4.06 (m, 2H). Anal. ($C_{17}H_{15}NO_4S$) C, H, N.

2-Bromo-1-phenylethanone (**33c**) (Acros) and 2-bromo-1-(3-methoxyphenyl)ethanone (**33d**) (Alfa Aesar) were commercially available, and 2-bromo-1-(3,4,5-trimethoxyphenyl)ethanone (**33e**) was prepared according to the literature.[40]

4-Hydroxybenzene-1,3-dicarbaldehyde (38). 38 was prepared from 4-hydroxybenzaldehyde according to the literature.[30]

3-[1,3]Dioxolan-2-ylbenzoic Acid (21c). 21c was prepared from 3-carboxybenzaldehyde (Aldrich) according to the analogous literature.[28] For details, see Supporting Information. ^{1}H NMR (DMSO-d_6): δ (ppm) = 13.09 (s, 1H), 8.00 (t, 1H, J = 1.6 Hz), 7.96 (d,t 1H, J_d = 7.7 Hz, J_t = 1.5 Hz), 7.68 (d,t, 1H, J_d = 7.7 Hz, J_t = 1.4 Hz), 7.53 (t, 1H, J = 7.7 Hz), 5.80 (s, 1H), 4.10−4.02 (m, 2H), 4.02−3.94 (m, 2H). Anal. ($C_{10}H_{10}O_4$) C, H.

Preparation of [1,3]Dioxolan-2-ylaroyl Chlorides 12 and 12c. The respective aroylic acid (20.0 mmol) was dissolved in dry THF and pyridine (60.0 mmol, 4.84 mL). $SOCl_2$ (40.0 mmol, 2.90 mL) was added, and the mixture was stirred for 30 min at room temperature. The precipitating pyridininium hydrochloride was filtered off. The solvent and excess reagent were removed in vacuo at room temperature, and the aroylic acid chlorides were purified by short-way column chromatography (SiO_2, CH_2Cl_2).

4-[1,3]Dioxolan-2-ylbenzoyl Chloride (12). ^{1}H NMR ($CDCl_3$): δ (ppm) = 8.13 (d, 2H, J = 8.5 Hz), 7.62 (d, 2H, J = 8.2 Hz), 5.88 (s, 1H), 4.12−4.10 (m, 2H), 4.08−4.06 (m,.2H). Anal. ($C_{10}H_9ClO_3$) C, H.

3-[1,3]Dioxolan-2-ylbenzoyl Chloride (12c). ^{1}H NMR ($CDCl_3$): δ (ppm) = 8.22 (t, 1H, J = 1.6 Hz), 8.11 (dt, 1H, J_d = 7.8 Hz, J_t = 1.6 Hz), 7.79 (d, 1H, J_d = 7.8 Hz), 7.53 (t, 1H, J = 7.8 Hz), 5.86 (s, 1H), 4.18−4.11 (m, 2H), 4.09−34.02 (m, 2H). Anal. ($C_{10}H_9ClO_3$) C, H.

Preparation of (1-Phenylsulfonyl-1*H*-2-indolyl)arylmethanones (13a−c and 25a,b) by Reaction of 1-Phenylsulfonyl-1*H*-indolyles with Carboxylic Acid Chlorides. The compounds were prepared according to the analogous literature.[25] For details, see Supporting Information.

(1-Benzenesulfonyl-1*H*-indol-2-yl)-(4-[1,3]dioxolan-2-ylphenyl)-methanone (13a). ^{1}H NMR (DMSO-d_6): δ (ppm) = 8.09−7.91 (m, 5H), 7.55−7.19 (m, 7H), 6.86 (d, 1H, J = 0.8 Hz), 5.84 (s, 1H), 4.07−4.04 (m, 2H), 4.02−3.99 (m, 2H). Anal. ($C_{24}H_{19}NO_5S$) C, H, N.

(1-Benzenesulfonyl-5-methoxy-1*H*-indol-2-yl)-(4-[1,3]dioxolan-2-ylphenyl)methanone (13b). ^{1}H NMR (DMSO-d_6): δ (ppm) = 7.95−7.87 (m, 5H), 7.74−7.58 (m, 5H), 7.24 (s, 1H), 7.19 (d, 1H, J = 2.5 Hz), 7.12 (dd, 1H, J = 9 Hz, J = 2.7 Hz), 5.87 (s, 1H), 4.08−4.05 (m, 2H), 4.03−3.97 (m, 2H), 3.77 (s, 3H). Anal. ($C_{25}H_{21}NO_6S$) C, H, N.

(1-Benzenesulfonyl-1*H*-indol-2-yl)-(3-[1,3]dioxolan-2-ylphenyl)-methanone (13c). ^{1}H NMR (DMSO-d_6): δ (ppm) = 8.08−7.34 (m, 13H), 7.31 (s, 1H), 5.86 (s, 1H), 4.08−4.05 (m, 2H), 4.0−3.97 (m, 2H). Anal. ($C_{24}H_{19}NO_5S$) C, H, N.

(1-Benzenesulfonyl-5-[1,3]dioxolan-2-yl-1*H*-indol-2-yl)phenyl-methanone (25a). 25a was used in the next step without further purification because of chemical instability.

(1-Benzenesulfonyl-5-[1,3]dioxolan-2-yl-1*H*-indol-2-yl)-(3-methoxyphenyl)methanone (25b). ^{1}H NMR (DMSO-d_6): δ (ppm) = 8.09 (d, 1H, J = 9.0 Hz), 7.81−7.70 (m, 2H), 7.69−7.41 (m, 6H), 7.37−7.30 (m, 2H), 5.82 (s, 1H), 4.10−3.93 (m, 6H), 3.84 (s, 3H). Anal. ($C_{25}H_{21}NO_6S \cdot 0.75H_2O$) C, H, N.

Preparation of (1-Benzenesulfonyl-1*H*-indole-2-carbonyl)-arylaldehydes (14a-c, 26a, and 26b) by Cleavage of the Respective [1,3]Dioxolanes. To a solution of the respective [1,3]dioxolanes (12.5 mmol) in CH_2Cl_2 (40 mL) was added $HClO_4$ (37%, 4.0 mL). The mixture was stirred at 0 °C for 1−2 h until TLC indicated complete consumption of the [1,3]dioxolane. The mixture was poured into water (100 mL), and the organic layer was separated, washed with saturated $NaHCO_3$ (50 mL), and dried (Na_2SO_4). The solvent was removed under reduced pressure, and the crude product so obtained was purified by column chromatography (SiO_2/CH_2Cl_2) and crystallization from diethyl ether.

4-(1-Benzenesulfonyl-1*H*-indole-2-carbonyl)benzaldehyde (14a). ^{1}H NMR ($CDCl_3$): δ (ppm) = 10.14 (s, 1H), 8.16−8.03 (m, 3H), 8.02−7.99 (m, 4H), 7.61−7.56 (m, 2H), 7.53−7.46 (m, 3H), 7.32 (t, 1H, J = 7.6 Hz), 7.02 (d, 1H, 0.5 Hz). Anal. ($C_{22}H_{15}NO_4S$) C, H, N.

4-(1-Benzenesulfonyl-5-methoxy-1*H*-indole-2-carbonyl)benzaldehyde (14b). ^{1}H NMR ($CDCl_3$): δ (ppm) = 10.13 (s, 1H), 8.10 (d, 2H, J = 8.2 Hz), 8.02 (t, 3H, J = 8.1 Hz), 7.92 (d, 2H, J = 7.9 Hz), 7.59−7.53 (m, 1H), 7.46 (t, 2H, J = 7.5 Hz), 7.10 (d,d, 1H, J = 2.73 Hz, J = 9.1 Hz), 6.98−6.96 (m, 2H), 3.82 (s, 3H). Anal. ($C_{23}H_{17}NO_5S$) C, H, N.

3-(1-Benzenesulfonyl-1*H*-indole-2-carbonyl)benzaldehyde (14c). ^{1}H NMR (DMSO-d_6): δ (ppm) = 10.13 (s, 1H), 8.40 (t, 1H, J = 1.5 Hz), 8.26−8.22 (m, 2H), 8.08 (d, 1H, J = 8.5 Hz), 7.98 (d, 2H, J = 7.1 Hz), 7.84 (t, 1H, J = 7.7 Hz), 7.76−7.71 (m, 2H), 7.63 (t, 2H, J = 7.5 Hz), 7.55 (t, 1H, J = 7.7 Hz), 7.40−7.36 (m, 2H). Anal. ($C_{22}H_{15}NO_4S$) C, H, N.

1-Benzenesulfonyl-2-benzoyl-1*H*-indole-5-carbaldehyde (26a). ^{1}H NMR (DMSO-d_6): δ (ppm) = 10.09 (s, 1H), 8.33−8.22 (m, 2H), 8.12−7.92 (m, 5H), 7.81−7.74 (m, 2H), 7.71−7.59 (m, 4H), 7.41 (s, 1H). Anal. ($C_{22}H_{15}NO_4S$) C, H, N.

1-Benzenesulfonyl-2-(3-methoxybenzoyl)-1*H*-indole-5-carbaldehyde (26b). ^{1}H NMR (DMSO-d_6): δ (ppm) = 10.07 (s, 1H), 8.33−8.25 (m, 2H), 8.11−8.02 (m, 3H), 7.82−7.64 (m, 3H), 7.55−7.44 (m, 4H), 7.38−7.32 (m, 1H), 3.82 (s, 3H). Anal. ($C_{23}H_{17}NO_5S$) C, H, N

Preparation of Acrylic Acid *tert*-Butyl Esters (15a−c, 27a,b, 35a,b, and 41a−c) by Wittig Olefination of the Respective Aldehydes. To a mixture of the pertinent aldehydes (12.0 mmol) were added (*tert*-butoxycarbonylmethyl)triphenylphosphonium chloride (Lancaster) (4.96 g, 12.0 mmol) in CH_2Cl_2 (100 mL), NaOH (0,98 g, 24mmol) in H_2O (50 mL), and NEt_3 (5 mL). The mixture was stirred at room temperature for 1 h. The organic layer was separated and washed with 1 M HCl (50 mL) and saturated aqueous $NaHCO_3$ (2 × 100 mL). The combined organic layers were dried (Na_2SO_4) and concentrated to give a crude product, which was purified by flash column chromatography (SiO_2, CH_2Cl_2) to afford the pure compound.

3-[4-(1-Benzenesulfonyl-1*H*-indole-2-carbonyl)phenyl]acrylic Acid *tert*-Butyl Ester (15a). ^{1}H NMR (DMSO-d_6): δ (ppm) = 8.05 (d, 1H, J = 9.0 Hz), 7.98−7.92 (m, 6H), 7.75−7.61 (m, 5H), 7.53 (t, 1H, J = 7.8 Hz), 7.39−7.34 (m, 2H), 6.72 (d, 1H, J = 16.2 Hz), 1.50 (s, 9H). Anal. ($C_{28}H_{25}NO_5S$) C, H, N.

3-[3-(1-Benzenesulfonyl-1*H*-indole-2-carbonyl)phenyl]acrylic Acid *tert*-Butyl Ester (15b). 15b was prepared as described above. The reaction time was 4 days. ^{1}H NMR (DMSO-d_6): δ (ppm) = 8.20 (s, 1H), 8.11−8.01 (m, 4H), 7.93 (d, 1H, J = 7.9 Hz), 7.83 (d, 2H, J = 7.8 Hz), 7.66−7.61 (m, 4H), 7.53 (t, 1H, J = 7.3 Hz), 7.39−7.30 (m, 2H), 6.63 (d, 1H, J = 16.2 Hz), 1.48 (s, 9H). Anal. ($C_{25}H_{19}NO_6S$) C, H, N.

3-[4-(1-Benzenesulfonyl-5-methoxy-1*H*-indole-2-carbonyl)-phenyl]acrylic Acid *tert*-Butyl Ester (15c). ^{1}H NMR (DMSO-d_6): δ (ppm) = 8.12−7.86 (m, 6H), 7.73−7.60 (m, 4H), 7.25−7.09 (m, 4H), 6.70 (d, 1H, J = 16.2 Hz), 3.77 (s, 3H), 1.49 (s, 9H). Anal. ($C_{25}H_{19}NO_6S$) C, H, N.

3-(1-Benzenesulfonyl-2-benzoyl-1*H*-indol-5-yl)acrylic Acid *tert*-Butyl Ester (27a). ^{1}H NMR (DMSO-d_6): δ (ppm) = 8.05−8.00 (m, 4H), 7.95−7.90 (m, 2H), 7.80−7.72 (m, 2H), 7.70−7.58 (m, 6H), 7.28 (s, 1H), 6.55 (d, 1H, J = 16.0 Hz), 1.48 (s, 9H). Anal. ($C_{28}H_{25}NO_5S$) C, H, N.

3-[1-Benzenesulfonyl-2-(3-methoxybenzoyl)-1*H*-indol-5-yl]-acrylic Acid *tert*-Butyl Ester (27b). ^{1}H NMR (DMSO-d_6): δ (ppm) = 8.10−8.00 (m, 4H), 7.95−7.85 (m, 1H), 7.80−7.25 (m, 10H), 6.55 (d, 1H, J = 16.2 Hz), 3.85 (s, 1H), 1.49 (s, 9H).

3-[4-(Benzofuran-2-carbonyl)phenyl]acrylic Acid *tert*-Butyl Ester (35a). ^{1}H NMR (DMSO-d_6): δ (ppm) = 8.02 (d, 2H, J = 8.2 Hz), 7.98 (d, 2H, J = 8.2 Hz), 7.87−7.85 (m, 2H), 7.81 (d, 1H, J = 8.5 Hz), 7.67 (d, 1H, J = 16.2 Hz), 7.59 (t, 1H, J = 7.8 Hz), 7.41 (t, 1H, J = 7.1 Hz), 6.72 (d, 1H, J = 16.2), 1.51 (s, 9H). Anal. ($C_{22}H_{20}O_4$) C, H.

3-[3-(Benzofuran-2-carbonyl)phenyl]acrylic Acid *tert*-Butyl Ester (35b). ^{1}H NMR (DMSO-d_6): δ (ppm) = 8.25 (s,1H), 8.07 (d, H, J = 7.9 Hz), 7.98 (d, 1H, J = 7.9 Hz), 7.90−7.85 (m, 2H), 7.80 (d, 1H, J = 8.5 Hz), 7.73−7.57 (m, 3H), 7.41 (t, 1H, J = 7.5 Hz), 6.67 (d, 1H, J = 16.2), 1.50 (m, 9H). Anal. ($C_{22}H_{20}O_4$) C, H.

3-(2-Benzoylbenzofuran-5-yl)acrylic Acid *tert*-Butyl Ester (41a). ^{1}H NMR (DMSO-d_6): δ (ppm) = 8.19−7.57 (m, 10H), 6.57 (d, 1H, J = 15.9 Hz), 1.49 (s, 9H). Anal. ($C_{22}H_{20}O_4$) C, H.

3-[2-(3-Methoxybenzoyl)benzofuran-5-yl]acrylic Acid *tert*-Butyl Ester (41b). ^{1}H NMR (DMSO-d_6): δ (ppm) = 8.13−7.24 (m, 9H), 6.55 (d, 1H, J = 16 Hz), 3.85 (s, 3H), 1.49 (s, 9H). Anal. ($C_{23}H_{22}O_5$) C, H.

3-[2-(3,4,5-Trimethoxybenzoyl)benzofuran-5-yl]acrylic Acid *tert*-Butyl Ester (41c). ^{1}H NMR (DMSO-d_6): δ (ppm) = 8.17−8.13 (m, 1H), 8.01−7.95 (m, 1H), 7.90−7.68 (m, 4H), 7.32 (s, 2H), 6.58 (d, 1H, J = 16.1 Hz), 3.89 (s, 6H), 3.80 (s, 3H), 1.50 (s, 9H). Anal. ($C_{25}H_{26}O_7$) C, H.

Preparation of the Acrylic Acids (16a−c, 28a,b, 36a,b, 42a−c) by Cleavage of the Respective Acrylic Acid *tert*-Butyl Esters. A solution of the respective acrylic acid *tert*-butyl ester (15.0 mmol) in 50 mL of TFA was stirred at room temperature for 1 h. The mixture was added to water (100 mL) with stirring, and the precipitating product was removed by filtration and dried in vacuum.

3-[4-(1-Benzenesulfonyl-1*H*-indole-2-carbonyl)phenyl]acrylic Acid (16a). ^{1}H NMR (DMSO-d_6): δ (ppm) = 8.05 (d, 1H, J = 9.0 Hz), 7.98−7.92 (m, 6H), 7.75−7.61 (m, 5H), 7.53 (t, 1H, J = 7.8 Hz), 7.45−7.31 (m, 2H), 6.72 (d, 1H, J = 16.2 Hz). Anal. ($C_{28}H_{25}NO_5S$) C, H, N.

3-[4-(1-Benzenesulfonyl-5-methoxy-1*H*-indole-2-carbonyl)-phenyl]acrylic Acid Monohydrate (16b). ^{1}H NMR (DMSO-d_6): δ (ppm) = 7.93−7.85 (m, 6H), 7.73−7.57 (m, 5H), 7.25 (s, 1H), 7.19 (d, 1H,J = 2.5 Hz), 7.14−7.11 (m 1H), 6.71 (d, 1H, J = 15.9 Hz), 3.77 (s, 3H).

3-[3-(1-Benzenesulfonyl-1*H*-indole-2-carbonyl)phenyl]acrylic Acid (16c). ^{1}H NMR (DMSO-d_6): δ (ppm) = 12.54 (s, 1H), 8.11 (s, 1H), 8.08 (t, 2H, J = 8.4 Hz), 7.41 (d, 2H, J = 7.4 Hz), 7.95 (d, 1H, J = 7.9 Hz), 7.76−7.71 (m 2H), 7.68−7.62 (m, 4H), 7.54 (t, 1H, J = 7.3 Hz), 7.39−7.35 (m, 2H), 6.64 (d, 1H, J = 16.2 Hz). Anal. ($C_{24}H_{17}NO_5S$) C, H, N.

3-(1-Benzenesulfonyl-2-benzoyl-1*H*-indol-5-yl)acrylic Acid (28a). ^{1}H NMR (DMSO-d_6): δ (ppm) = 12.45 (s, br, 1H), 8.09−7.85 (m, 7H), 7.79−7.58 (m, 7H), 7.29 (s, 1H), 6.55 (d, 1H, J = 16.2 Hz). Anal. ($C_{24}H_{17}NO_5S\cdot0.25H_2O$) C, H, N.

3-[1-Benzenesulfonyl-2-(3-methoxybenzoyl)-1*H*-indol-5-yl]-acrylic Acid (28b). ^{1}H NMR (DMSO-d_6): δ (ppm) = 12.30 (s, br, 1H), 8.10−8.02 (m, 4H), 7.92−7.87 (m, 1H), 7.78−7.63 (m, 4H), 7.55−7.41 (m, 3H), 7.37−7.29 (m, 2H), 6.55 (d, 1H, J = 15.9 Hz), 3.85 (s, 3H). Anal. ($C_{25}H_{19}NO_6S\cdot0.75H_2O$) C, H, N.

3-[4-(Benzofuran-2-carbonyl)phenyl]acrylic Acid (36a). ^{1}H NMR (DMSO-d_6): δ (ppm) = 12.63 (s, 1H), 8.03 (d, 2H, J = 8.5 Hz), 7.93 (d, 2H, J = 8.5 Hz), 7.89−7.85 (m, 2H), 7.79 (d, 1H, J = 8.5 Hz), 7.71 (d, 1H, J = 16.2 Hz), 7.59 (t, 1H, J = 7.1 Hz), 7.41 (t, 1H, J = 7.1 Hz), 6.72 (d, 1H, J = 16.1 Hz). Anal. ($C_{18}H_{12}O_4$) C, H.

3-[3-(Benzofuran-2-carbonyl)phenyl]acrylic Acid (36b). ^{1}H NMR (DMSO-d_6): δ (ppm) = 12.55 (s, 1H), 8.24 (s, 1H), 8.07 (d, 1H, J = 7.7 Hz), 8.00 (d, 1H, J = 7.3 Hz), 7.90−7.85 (m, 2H), 7.80 (d, 1H, J = 8.5 Hz), 7.74 (d, 1H, J = 16.2 Hz), 7.66 (t, 1H, J = 7.7 Hz), 7.60 (t, 1H, J = 7.8 Hz), 7.41 (t, 1H, J = 7.5 Hz), 6.69 (d, 1H, J = 16.1 Hz). Anal. ($C_{18}H_{12}O_4$) C, H.

3-(2-Benzoylbenzofuran-5-yl)acrylic Acid (42a). ^{1}H NMR (DMSO-d_6): δ (ppm) = 12.45 (s, 1H), 8.20−7.58 (m, 11H), 6.58 (d, 1H, J = 15.9 Hz). Anal. ($C_{18}H_{12}O_4$) C, H.

3-[2-(3-Methoxybenzoyl)benzofuran-5-yl]acrylic Acid (42b). ^{1}H NMR (DMSO-d_6): δ (ppm) = 12.50 (s, br, 1H), 8.17−8.14 (m, 1H), 8.02−7.94 (m, 1H), 7.88−7.78 (m, 2H), 7.74 (d, 1H, J = 15.9 Hz), 7.62−7.47 (m, 3H), 7.35−7.28 (m, 1H), 6.59 (d, 1H, J = 15.9 Hz), 3.87 (s, 3H). Anal. ($C_{19}H_{14}O_5 \cdot 0.25H_2O$) C, H.

3-[2-(3,4,5-Trimethoxybenzoyl)benzofuran-5-yl]acrylic Acid (42c). ^{1}H NMR (DMSO-d_6): δ (ppm) = 12.45 (s, br, 1H), 8.17−8.13 (m, 1H), 7.99−7.03 (m, 1H), 7.90−7.81 (m, 2H), 7.75 (d, 1H, J = 15.9 Hz), 7.31 (s, 2H), 6.58 (d, 1H, J = 15.9 Hz), 3.89 (s, 6H), 3.80 (s, 3H). Anal. ($C_{21}H_{18}O_7 \cdot 0.25H_2O$) C, H.

Preparation of 3-[4-(1*H*-Indole-2-carbonyl)phenyl]acrylic Acids by Cleavage of the Benzenesulfonyl Group. The N-protected methanone derivative **X** (1.5 mmol) was heated in methanol (50 mL) and 10% aqueous NaOH (50 mL) under reflux for 12 h. The organic solvent was removed under reduced pressure, and the aqueous solution was acidified (HCl, 37%). The yellow precipitating crystals were removed by filtration, washed with diluted HCl, and dried in vacuo. Thus, compounds **17a**−**c** and **29a**,**b** were prepared.

3-[4-(1*H*-Indole-2-carbonyl)phenyl]acrylic Acid (17a). ^{1}H NMR (DMSO-d_6): δ (ppm) = 12.02 (s, 1H), 7.92 (d, 2H, J = 8.2 Hz), 7.72 (d, 3H, J = 8.2 Hz), 7.51 (d, 1H, J = 8.0 Hz), 7.34−7.24 (m, 2H), 7.18 (d, 1H, J = 1.4 Hz), 7.10 (t, 1H, J = 7.5 Hz), 6.61 (d, 1H, J = 15.9 Hz). Anal. ($C_{18}H_{13}NO_3 \cdot H_2O$) C, H, N.

3-[4-(5-Methoxy-1*H*-indole-2-carbonyl)phenyl]acrylic Acid (17b). ^{1}H NMR (DMSO-d_6): δ (ppm) = 12.59 (s, 1H, exchangeable), 11.89 (d, 1H, J = 1.6 Hz, exchangeable), 7.94 (d, 2H, J = 7.9 Hz), 7.90 (d, 2H, J = 7.9 Hz), 7.70 (d, 1H, J = 16.0 Hz), 7.40 (d, 1H, 8.8 Hz), 7.16 (d, 1H, J = 2.5 Hz), 7.06 (d, 1H, J = 1.6 Hz), 6.99 (dd, 1H, J = 8.8 Hz, J = 2.5 Hz), 6.70 (d, 1H, J = 16.0 Hz), 3.77 (s, 3H). Anal. ($C_{19}H_{15}NO_4 \cdot {}^1/_6H_2O$) C, H, N.

3-[3-(1*H*-Indole-2-carbonyl)phenyl]acrylic Acid Hydrate (17c). ^{1}H NMR (DMSO-d_6): δ (ppm) = 12.18 (s, 1H), 8.00 (s, 1H), 7.85−7.81 (m,2H), 7.74 (d, 1H, J = 8.2 Hz), 7.56 (t, 2H, J = 8.1 Hz), 7.35−7.30 (m, 2H), 7.15−7.07 (m, 2H), 6.68 (d, 1H, J = 15.9). Anal. ($C_{18}H_{13}NO_3 \cdot {}^3/_2H_2O$) C, H, N.

3-(2-Benzoyl-1*H*-indol-5-yl)acrylic Acid (29a). ^{1}H NMR (DMSO-d_6): δ (ppm) = 12.27 (s, br, 1H), 12.22 (s, 1H), 8.04−7.92 (m, 3H), 7.75−7.50 (m, 6H), 7.20−7.17 (m, 1H), 6.45 (d, 1H, J = 15.9 Hz). Anal. ($C_{18}H_{13}NO_3 \cdot 0.5H_2O$) C, H, N.

3-[2-(3-Methoxybenzoyl)-1*H*-indol-5-yl]acrylic Acid (29b). ^{1}H NMR (DMSO-d_6): δ (ppm) = 12.30 (s, br, 1H), 12.21 (s, 1H), 8.07 (s, 1H), 7.75−7.49 (m, 6H), 7.43−7.40 (m, 1H), 7.30−7-23 (m, 1H), 7.21−7.18 (m, 1H), 6.45 (d, 1H, J = 15.9 Hz), 3.86 (s, 3H). Anal. ($C_{19}H_{15}NO_4 \cdot 0.5H_2O$) C, H, N.

Preparation of *N*-(Tetrahydropyran-2-yloxy)acrylamides by Amidation of the Respective Acrylic Acids (18a−c, 19a−c, 30a,b, 31a,b, 37a,b, 43a−c). To a solution of the respective acrylic acid (7.1 mmol), BOP {(benzotriazol-1-yloxy)tris(dimethylamino)-phosphonium hexafluorophosphate} (3.32 g, 7.5 mmol), and NEt_3 (2.3 mL, 17.04 mmol) in dry THF or DMF (30 mL) was added NH_2OTHP {*O*-(tetrahydropyran-2-yl)hydroxylamine, Aldrich} (1.0 g, 8.5 mmol). The mixture was stirred for 1−2 h (TLC control) and then poured into water (100 mL) with stirring. The precipitating crude product was removed by filtration and dried in vacuo. Purification by column chromatography (SiO_2, ethyl acetate) afforded the pure title product as a colorless solid.

3-[4-(1-Benzenesulfonyl-1*H*-indole-2-carbonyl)phenyl]-*N*-(tetrahydropyran-2-yloxy)acrylamide (18a). ^{1}H NMR (DMSO-d_6): δ (ppm) = 11.40 (s, 1H, exchangeable), 8.06 (d, 1H, J = 8.4 Hz), 7.98−7.93 (m, 4H), 7.79 (d, 2H, J = 8.5 Hz), 7.73−7.70 (m, 2H), 7.66−7.49 (m, 4H), 7.39−7.33 (m, 2H), 6.68 (d, 1H, J = 15.9 Hz), 4.94 (s, 1H), 3.97 (m, br, 1H), 3.55 (d, br, 1H, J = 11.0 Hz), 1.71 (s, br, 3H), 1.55 (s, br, 3H). Anal. ($C_{29}H_{26}N_2O_6S$) C, H, N.

3-[4-(1-Benzenesulfonyl-5-methoxy-1*H*-indole-2-carbonyl)-phenyl]-*N*-(tetrahydropyran-2-yloxy)acrylamide (18b). ^{1}H NMR (DMSO-d_6): δ (ppm) = 11.40 (s, 1H), 7.96−7.88 (m, 5H), 7.79 (d, 2H, J = 8.3 Hz), 7.72 (t, 1H, J = 7.4 Hz), 7.63−7.58 (m 3H), 7.25 (s, 1H), 7.19 (d, 1H, J = 2.5 Hz), 7.12 (d,d, 1H, J = 2.5 Hz, J = 9.2 Hz), 6.68 (d, 1H, J = 15.9 Hz), 4.95 (s, 3H), 3.97 (m, 1H), 3.57−3.53 (m, 1H), 1.71 (m, 3H), 1.55 (m, 3H). Anal. ($C_{30}H_{28}N_2O_7S$) C, H, N.

3-[3-(1-Benzenesulfonyl-1*H*-indole-2-carbonyl)phenyl]-*N*-(tetrahydropyran-2-yloxy)acrylamide (18c). ^{1}H NMR (DMSO-d_6): δ (ppm) = 11.27 (s, 1H), 8.13−8.01 (m, 4H), 7.96−7.90 (m, 2H), 7.77−7.72 (m, 2H), 7.68−7.62 (m 4H), 7.58−7.52 (m, 1H), 7.40−7.35 (m, 2H), 6.62 (d, 1H, J = 15.9 Hz), 4.92 (s, 1H), 3.04−3.94 (m, 1H), 3.55−3.52 (m, 1H), 1.69 (m, 3H), 1.54 (m, 3H). Anal. ($C_{29}H_{26}N_2O_6$) C, H, N.

3-[4-(1*H*-Indole-2-carbonyl)phenyl]-*N*-(tetrahydropyran-2-yloxy)acrylamide (19a). ^{1}H NMR (DMSO-d_6): δ (ppm) = 12.02 (s, 1H), 11.38 (s, 1H), 7.98 (d, 2H, J = 8.2 Hz), 7.79 (d, 2H, J = 8.2 Hz), 7.73 (d, 1H, J = 7.9 Hz), 7.60 (d, 1H, J = 15.6 Hz), 7.250 (d, 1H, J = 8.5 Hz), 7.33 (t, 1H, J = 7.6 Hz), 7.18 (d, 1H, J = 1.4 Hz), 7.11 (t, 1H, J = 7.5 Hz), 6.67 (d, 1H, J = 15.6 Hz), 4.95 (s, 1H), 3.97 (m, 1H), 3.57−3.54 (m, 1H), 1.71 (m, 3H), 1.55 (m, 3H). Anal. ($C_{23}H_{22}N_2O_4$) C, H, N.

3-[4-(5-Methoxy-1*H*-indole-2-carbonyl)phenyl]-*N*-(tetrahydropyran-2-yloxy)acrylamide (19b). ^{1}H NMR (DMSO-d_6): δ (ppm) = 11.89 (s, 1H), 11.37 (s, 1H), 7.96 (d, 2H, J = 8.3 Hz), 7.78 (d, 2H, J = 8.2 Hz), 7.60 (d, 1H, J = 16.2 Hz), 7.40 (d 1H, J = 8.8 Hz), 7.15 (d, 1H, J = 2.2 Hz), 7.16 (d, 1H, J = 2.2 Hz), 7.04 (d,d, 1H, J = 2.5 Hz, J = 9.1 Hz), 6.67 (d, 1H, J = 15.9 Hz), 4.94 (s, 1H), 4.00−3.97 (m, 1H), 3.86 (s, 3H), 3.63−3.57 (m, 1H), 1.71 (m, 3H), 1.55 (m, 3H). Anal. ($C_{24}H_{24}N_2O_5$) C, H, N.

3-[3-(1*H*-Indole-2-carbonyl)phenyl]-*N*-(tetrahydropyran-2-yloxy)acrylamide (19c). ^{1}H NMR (DMSO-d_6): δ (ppm) = 12.04 (s, 1H), 11.27 (s, 1H), 8.10 (s, 1H), 7.92 (t, 2H, J = 9.3 Hz), 7.74 (d, 1H, J = 8.0 Hz), 7.67−7.60 (m, 2H), 7.51 (d, 1H, J = 8.2 Hz), 7.33 (t, 1H, J = 7.7 Hz), 7.18 (d, 1H, J = 1.4 Hz), 7.11 (t, 1H, J = 7.1 Hz), 6.65 (d, 1H, J = 15.7 Hz), 4.93 (s, 1H), 3.95 (m, 1H), 3.56−3.52 (m, 1H), 1.70 (m, 3H), 1.54 (m, 3H). Anal. ($C_{23}H_{22}N_2O_4$) C, H, N.

3-(2-Benzoyl-1*H*-indol-5-yl)-*N*-(tetrahydropyran-2-yloxy)-acrylamide (30a). ^{1}H NMR (DMSO-d_6): δ (ppm) = 12.20 (s, br, 1H), 11.18 (s, br, 1H), 7.96−7.93 (m, 3H), 7.74−7.67 (m, 1H), 7.63−7.52 (m, 5H), 7.18 (s, 1H), 6.45 (d, 1H, J = 15.4 Hz), 4.91 (s, 1H), 4.10−3.90 (m, 1H), 3.58−3.50 (m, 1H), 1.77−1.63 (m, 3H), 1.63−1.48 (m, 3H). HR-PI-LSI-MS calcd for $C_{23}H_{23}N_2O_4$ [$MH^{+\bullet}$]: 391.1658. Found 391.1669.

3-[1-Benzenesulfonyl-2-(3-methoxybenzoyl)-1*H*-indol-5-yl]-*N*-(tetrahydropyran-2-yloxy)acrylamide (30b). ^{1}H NMR (DMSO-d_6): δ (ppm) = 11.25 (s, br, 1H), 8.12−7.90 (m, 3H), 7.95−7.88 (m, 1H), 7.78−7.72 (m, 2H), 7.70−7.46 (m, 5H), 7.45−7.41 (m, 1H), 7.37−7.30 (m, 2H), 6.53 (d, 1H, J = 15.7 Hz), 4.92 (s, br, 1H), 4.03−3.90 (m, 1H), 3.83 (s, 3H), 3.60−3.50 (m, 1H), 1.75−1.45 (m, 6H). Anal. ($C_{30}H_{28}N_2O_7S$) C, H, N.

3-(2-Benzoyl-1*H*-indol-5-yl)-*N*-(tetrahydropyran-2-yloxy)-acrylamide (31a). ^{1}H NMR (DMSO-d_6): δ (ppm) = 12.20 (s, br, 1H), 11.18 (s, br, 1H), 7.96−7.93(m, 3H), 7.73−7.68 (m, 1H), 7.62−7.52 (m, 5H), 7.19 (s, 1H), 6.45 (d, 1H, J = 15.4 Hz), 4.91 (s, 1H), 4.01−3.91 (m, 1H), 3.56−3.52 (m, 1H), 1.71−1.54 (m, 6H). IR (KBr): ν (cm^{-1}) = 3290, 2948, 1652, 1625. HR-PI-EI-MS calcd for $C_{23}H_{22}N_2O_4$ [$M^{+\bullet}$]: 390.1580. Found 390.1580.

3-[2-(3-Methoxybenzoyl)-1*H*-indol-5-yl]-*N*-(tetrahydropyran-2-yloxy)acrylamide (31b). ^{1}H NMR (DMSO-d_6): δ (ppm) = 12.20 (s, br, 1H), 11.18 (s, br, 1H), 7.93 (s, 1H), 7.63−7.50 (m, 5H), 7.43−7.38 (m, 1H), 7.30−7.24 (m, 1H), 7.20 (s, 1H), 6.45 (d, 1H, J = 15.6 Hz), 4.91 (s, 1H), 4.03−3.90 (m, 1H), 3.85 (s, 3H), 3.60−3.50 (m, 1H), 1.78−1.47 (m, 6H). Anal. ($C_{24}H_{24}N_2O_5 \cdot 0.5H_2O$) C, H, N.

3-[4-(Benzofuran-2-carbonyl)phenyl]-*N*-(tetrahydropyran-2-yloxy)acrylamide (37a). ^{1}H NMR (DMSO-d_6): δ (ppm) = 11.39 (s, 1H), 8.06 (d, 2H, J = 8.2 Hz), 7.89−7.78 (m, 5H), 7.41 (t, 1H, J = 7.5 Hz), 6.69 (d, 1H, J = 15.9 Hz), 4.95 (s, 1H), 3.99 (m, 1H), 3.58−3.54 (m, 1H), 1.72 (m, 3H), 1.56 (m, 3H). Anal. ($C_{23}H_{21}NO_5 \cdot {}^1/_4H_2O$) C, H, N.

3-[3-(Benzofuran-2-carbonyl)phenyl]-*N*-(tetrahydropyran-2-yloxy)acrylamide (37b). ^{1}H NMR (DMSO-d_6): δ (ppm) = 11.29 (s, 1H), 8.16 (s, 1H), 7.99 (d, 1H, J = 7.7 Hz), 7.93 (d, 1H, J =

8.0 Hz), 7.89−7.86 (m, 2H), 7.80 (d, 1H, J = 8.5 Hz), 7.68−7.57 (m, 3H), 7.41 (t, 1H, J = 7.5 Hz), 6.66 (d, 1H, J = 15.9 Hz), 4.93 (s, 1H), 3.96 (m, 1H), 3.56−3.52 (m, 1H), 1.70 (m, 3H), 1.54 (m, 3H). Anal. ($C_{23}H_{21}NO_5$) C, H, N.

3-(2-Benzoylbenzofuran-5-yl)-*N*-(tetrahydropyran-2-yloxy)-acrylamide (43a). ^{1}H NMR (DMSO-d_6): δ (ppm) = 11.29 (s, 1H), 8.04−7.97 (m, 3H), 7.85−7.81 (m, 3H), 7.69−7.58(m, 4H), 6.55 (d, 1H, J = 15.7 Hz), 4.93 (s, 1H), 3.66−3.46 (m, 1H), 1.78−1.47 (m, 7H). Anal. ($C_{23}H_{21}NO_5$) C, H, N.

3-[2-(3-Methoxybenzoyl)benzofuran-5-yl]-*N*-(tetrahydropyran-2-yloxy)acrylamide (43b). ^{1}H NMR (DMSO-d_6): δ (ppm) = 11.30 (s, br, 1H), 8.07 (s, 1H), 7.87−7.81 (m, 3H), 7.63−7.47 (m, 4H), 7.34−7.28 (m, 1H), 6.55 (d, 1H, J = 15.6 Hz), 4.93 (s, 1H), 4.05−3.92 (m, 1H), 3.86 (s, 3H), 3.60−3.52 (m, 1H), 1.78−1.45 (m, 6H). Anal. ($C_{24}H_{23}NO_6 \cdot {}^1/_3H_2O$) C, H, N.

***N*-(Tetrahydropyran-2-yloxy)-3-[2-(3,4,5-trimethoxybenzoyl)-benzofuran-5-yl]acrylamide (43c).** ^{1}H NMR (DMSO-d_6): δ (ppm) = 11.28 (s, br, 1H), 8.05 (s, br, 1H), 7.91 (s, 1H), 7.88−7.77 (m, 2H), 7.63 (d, 1H, J = 15.8 Hz), 7.31 (s, 2H), 6.55 (d, 1H, J = 15.8 Hz), 4.93 (s, br, 1H), 4.05−3.90 (m, 1H), 3.89 (s, 6H), 3.80 (s, 3H), 3.59−3.51 (m, 1H), 1.78−1.47 (m, 6H). Anal. ($C_{26}H_{27}NO_8 \cdot 1.5H_2O$) C, H, N.

Synthesis of *N*-Hydroxyacrylamides by Cleavage of *N*-(Tetrahydropyran-2-yloxy)acrylamides (5a−c, 6a−c, 7a,b, 8a,b, 9a,b, 10a−c). The respective *N*-(tetrahydropyran-2-yloxy)acrylamides (0.25 mmol) were dissolved in MeOH (20.0 mL). HCl (0.6 N, 20.0 mL) was added, and the mixture was stirred over night at room temperature. The precipitating crystals were filtered off, washed with HCl (0.6 N), and dried in vacuo.

3-[4-(1-Benzenesulfonyl-1*H*-indole-2-carbonyl)phenyl]-*N*-hydroxyacrylamide Semihydrate (5a). Yield: 1.76 g (80%). Mp: 149.7−151.3 °C. ^{1}H NMR (DMSO-d_6): δ (ppm) = 10.91 (s, 1H), 9.17 (s, 1H), 8.05 (d, 1H, J = 8.5 Hz), 7.99−7.93 (m, 6H), 7.79−7.59 (m, 5H), 7.53 (t, 1H, J = 7.8 Hz), 7.39−7.33 (m, 2H), 6.63 (d, 1H, J = 15.9 Hz). Anal. ($C_{24}H_{18}N_2O_6S \cdot 0.5H_2O$) C, H, N. IR (KBr): ν (cm^{-1}) = 1659, 1627. EI-MS (70 eV) *m*/*z* (%): 446 (4.8) $[M]^{+\bullet}$, 77 (100).

3-[4-(1-Benzenesulfonyl-5-methoxy-1*H*-indole-2-carbonyl)-phenyl]-*N*-hydroxyacrylamide (5b). Yield: 0.47 g (56%). Mp: 141.8−142.0 °C. ^{1}H NMR (DMSO-d_6): δ (ppm) = 10.92 (s, 1H), 9.17 (s,1H), 7.96−7.88 (m, 6H), 7.79−7.69 (m, 3H), 7.61 (t, 2H, J = 7.5 Hz), 7.24 (s, 1H), 7.19 (d, 1H, J = 7.2 Hz), 7.12 (d,d, 1H, J = 2.7 Hz, J = 9.0 Hz), 3.77 (s, 3H). Anal. ($C_{25}H_{20}N_2O_6S$) C, H, N. IR (KBr): ν (cm^{-1}) = 1659, 726. ES-MS (CH_2Cl_2/MeOH + 10 mmol/L NH_4Ac) *m*/*z* (%): 477 (100) $[M + H^+]^+$.

3-[3-(1-Benzenesulfonyl-1*H*-indole-2-carbonyl)phenyl]-*N*-hydroxyacrylamide Semihydrate (5c). Yield: 0.27 g (73%). Mp: 123.0−129.1 °C. ^{1}H NMR (DMSO-d_6): δ (ppm) = 10.78 (s, 1H), 9.12 (s, 1H), 8.11−8.00 (m, 4H), 7.95−7.88 (m, 2H), 7.77−7.72 (m, 2H), 7.67−7.60 (m 3H), 7.56−7.52 (m, 3H), 7.40−7.35 (m, 2H), 6.57 (d, 1H, J = 15.9 Hz), 4.95 (s, 1H), 3.97 (m, 1H), 3.57−3.53 (m, 1H), 1.71 (m, 3H), 1.55 (m, 3H). Anal. ($C_{24}H_{18}N_2O_5S \cdot {}^1/_2H_2O$) C, H, N. IR (KBr): ν (cm^{-1}) = 3214, 1665, 1596. ES-MS (CH_2Cl_2/MeOH + 10 mmol/L NH_4Ac) *m*/*z* (%): 464 (100) $[M + NH_4^+]^+$, 447 (77) $[M + H^+]^+$.

***N*-Hydroxy-3-[4-(1*H*-indole-2-carbonyl)phenyl]acrylamide (6a).** Yield: 0.04 g (85%). Mp: 183.9−192.8 °C. ^{1}H NMR (DMSO-d_6): δ (ppm) = 10.92 (s, 1H), 7.98 (d, 2H, J = 8.5 Hz), 7.77 (d, 2H, J = 8.5 Hz), 7.73 (d, 1H, J = 8.2 Hz), 7.57 (d 1H, J = 15.9 Hz), 7.51 (d, 1H, J = 7.5 Hz), 7.32 (t, 1H, J = 7.7 Hz), 7.18 (d, 1H, J = 1.4 Hz), 7.09 (t,1H, J = 7.3 Hz), 6.60 (d, 1H, J = 15.9). Anal. ($C_{18}H_{14}N_2O_3$) C, H, N. IR (KBr): ν (cm^{-1}) = 1617, 1520. ES-MS (CH_2Cl_2/MeOH + 10 mmol/L NH_4Ac) *m*/*z* (%): 307 (100) $[M + H^+]^+$.

***N*-Hydroxy-3-[4-(5-methoxy-1*H*-indole-2-carbonyl)phenyl]-acrylamide Semihydrate (6b).** Yield: 0.06 g (75%). Mp: 204.2−204.3 °C. ^{1}H NMR (DMSO-d_6): δ (ppm) = 11.88 (s, 1H), 10.89 (s, 1H), 9.17 (s, 1H), 7.9 (d, 2H, J = 8.2 Hz), 7.77 (d, 2H, J = 8.2 Hz), 7.57 (d, 1H, J = 15.9 Hz), 7.40 (d, 1H, J = 9.0 Hz), 7.15 (d, 1H, J = 2.2 Hz), 7.07 (d, 1H, J = 1.4 Hz), 6.99 (dd, 1H, J = 2.5 Hz, J = 9.1 Hz), 6.62 (d, 1H, J = 15.7), 3.76 (s, 3H). Anal. ($C_{19}H_{16}N_2O_4 \cdot {}^1/_2H_2O$) C, H, N. IR (KBr): ν (cm^{-1}) = 3264, 1679, 1653, 1611. ES-MS (CH_2Cl_2/MeOH + 10 mmol/L NH_4Ac) *m*/*z* (%): 336 (21) $[M]^+$, 335 (100).

***N*-Hydroxy-3-[3-(1*H*-indole-2-carbonyl)phenyl]acrylamide Monohydrate (6c).** Yield: 0.14 g (96%). Mp: 190.6−203.2 °C. ^{1}H NMR (DMSO-d_6): δ (ppm) = 12.04 (s, 1H), 10.80 (s, 1H), 9.13 (s, 1H), 8.08 (s, 1H), 7.94−7.87 (m, 2H), 7.74 (d, 1H, J = 8.0 Hz), 7.64 (t, 1H, J = 7.7 Hz), 7.60 (d, 1H, J = 15.6 Hz), 7.52 (d, 1H, J = 8.2 Hz), 7.33 (d, 1H, J = 7.1 Hz), 7.18 (d, 1H, J = 1.4 Hz), 7.11 (t, 1H J = 7.1 Hz), 6.60 (d, 1H, J = 15.6 Hz). Anal. ($C_{18}H_{14}N_2O_3$) C, H, N. IR (KBr): ν (cm^{-1}) = 1659, 1624. ES-MS (CH_2Cl_2/MeOH + 10 mmol/L NH_4Ac) *m*/*z* (%): 307 (100) $[M + H^+]^+$.

3-[4-(Benzofuran-2-carbonyl)phenyl]-*N*-hydroxyacrylamide (7a). Yield: 0.17 g (89%). Mp: 177 °C (dec). ^{1}H NMR (DMSO-d_6): δ (ppm) = 10.93 (s, 1H), 8.04 (d, 2H, J = 8.5 Hz), 7.88−7.77 (m, 5H), 7.61−7.55 (m, 2H), 7.41 (t, 1H, J = 7.5 Hz), 6.65 (d, 1H, J = 15.9 Hz). Anal. ($C_{18}H_{13}NO_4 \cdot {}^1/_4H_2O$) C, H, N. IR (KBr): ν (cm^{-1}) = 1653, 697. ES-MS (CH_2Cl_2/MeOH + 10 mmol/L NH_4Ac) *m*/*z* (%): 308 (100) $[M + H^+]^+$.

3-[3-(Benzofuran-2-carbonyl)phenyl]-*N*-hydroxyacrylamide (7b). Yield: 0.27 g (98%). Mp: 170.7 °C. ^{1}H NMR (DMSO-d_6): δ (ppm) = 10.81 (s, 1H), 9.15 (s, 1H), 8.15 (s, 1H), 7.97 (d, 1H, J = 7.7 Hz), 7.93−7.85 (m, 3H), 7.80 (d, 1H, J = 8.2 Hz), 7.67−7.57 (m, 3H), 7.41 (t, 1H, J = 7.5 Hz), 6.60 (d, 1H, J = 15.6 Hz). Anal. ($C_{18}H_{13}NO_4$) C, H, N. IR (KBr): ν (cm^{-1}) = 1642, 761. ES-MS (CH_2Cl_2/MeOH + 10 mmol/L NH_4Ac) *m*/*z* (%): 615 (100) $[2M + H]^+$, 325 (31) $[M + NH_4^+]^+$, 308 (100) $[M + H]^+$.

3-(1-Benzenesulfonyl-2-benzoyl-1*H*-indol-5-yl)-*N*-hydroxyacrylamide (8a). Yield: 0.48 g (77%). Mp: 170−172 °C. ^{1}H NMR (DMSO-d_6): δ (ppm) = 10.79 (s, br, 1H), 9.05 (s, br, 1H), 8.10−7.85 (m, 6H), 7.80−7.50 (m, 8H), 7.31 (s, 1H), 6.45 (d, 1H, J = 15.9 Hz). IR (KBr): ν (cm^{-1}) = 2950, 1664, 1630. EI-MS *m*/*z* (%): 446 (25) $[M]^{+\bullet}$, 428 (80), 287 (75), 105 (100). HR-PI-EI-MS calcd for $C_{24}H_{18}N_2O_5S$ $[M^{+\bullet}]$: 446.0936. Found 446.0929.

3-[1-Benzenesulfonyl-2-(3-methoxybenzoyl)-1*H*-indol-5-yl]-*N*-hydroxyacrylamide (8b). Yield: 0.06 g of colorless crystals (43%). Mp: 135−138 °C. ^{1}H NMR (DMSO-d_6): δ (ppm) = 10.79 (s, br, 1H), 9.06 (s, br, 1H), 8.12−7.97 (m, 3H), 7.89 (s, 1H), 7.80−7.42 (m, 8H), 7.35−7.28 (m, 2H), 6.48 (d, 1H, J = 15.9 Hz), 3.84 (s, 3H). IR (KBr): ν (cm^{-1}) = 2833, 1660. PI-LSI-MS *m*/*z* (%): 477 (90) $[MH^{+\bullet}]$. HR-PI-LSI-MS calcd for $C_{25}H_{21}N_2O_6S$ $[MH]^{+\bullet}$: 477.1120. Found 477.1118.

3-(2-Benzoyl-1*H*-indol-5-yl)-*N*-hydroxyacrylamide (9a). Yield: 0.13 g of yellow crystals (53%). Mp: 214−216 °C. ^{1}H NMR (DMSO-d_6): δ (ppm) = 12.19 (s, br, 1H), 10.69 (s, br, 1H), 9.00 (s, br, 1H), 7.98−7.87 (m, 3H), 7.73−7.50 (m, 6H), 7.18 (s, 1H), 6.40 (d, 1H, J = 15.7 Hz). Anal. ($C_{18}H_{14}N_2O_3$) C, H, N. IR (KBr): ν (cm^{-1}) = 3322, 1610. ESI-MS *m*/*z* (%): 307 (100) $[MH]^{+\bullet}$.

***N*-Hydroxy-3-[2-(3-methoxybenzoyl)-1*H*-indol-5-yl]acrylamide (9b).** Yield: 0.03 g of yellow crystals (53%). Mp: 155−156 °C. ^{1}H NMR (DMSO-d_6): δ (ppm) = 12.17 (s, br, 1H), 10.70 (s, br, 1H), 8.99 (s, br, 1H), 7.91 (s, 1H), 7.62−7.47 (m, 5H), 7.44−7.39 (m, 1H), 7.32−7.23 (m, 1H), 7.22−7.18 (m, 1H), 6.40 (d, 1H, J = 15.7 Hz), 3.86 (s, 3H). Anal. ($C_{19}H_{16}N_2O_4 \cdot 0.5H_2O$) C, H, N. IR (KBr): ν (cm^{-1}) = 3311, 1613. ESI-MS *m*/*z* (%): 673 (45) $[2MH]^{+\bullet}$, 337 (100) $[MH]^{+\bullet}$.

3-(2-Benzoylbenzofuran-5-yl)-*N*-hydroxyacrylamide (10a). Crystallization from ethanol/H_2O (1:1) afforded 0.03 g (53%) of colorless crystals. Mp: 151 °C. ^{1}H NMR (DMSO-d_6): δ (ppm) = 10.80 (s, 1H), 9.07 (s, 1H), 8.06−7.97 (m, 3H), 7.86−7.79 (m, 3H), 7.73 (d, 1H, J = 7.4 Hz), 7.66−7.62 (m, 2H), 7.59 (d, 1H, J = 6.9 Hz), 6.50 (d, 1H, J = 15.8 Hz). Anal. ($C_{18}H_{13}NO_4 \cdot 0.5EtOH$) C, H, N. IR (KBr): ν (cm^{-1}) = 3578, 3431, 1645. ES-MS (CH_2Cl_2/MeOH/CH_3COONH_4) *m*/*z* (%): 306 (100) $[M - H^+]^-$.

***N*-Hydroxy-3-[2-(3-methoxybenzoyl)benzofuran-5-yl]acrylamide (10b).** Yield: 0.75 g colorless crystals (55%). Mp: 138−140 °C. ^{1}H NMR (DMSO-d_6): δ (ppm) = 10.80 (s, br, 1H), 9.12 (s, br, 1H), 8.05 (s, 1H), 7.88−7.76 (m, 3H), 7.65−7.47 (m, 4H), 7.33−7.27 (m, 1H), 6.50 (d, 1H, J = 15.9 Hz). Anal. ($C_{19}H_{15}NO_5 \cdot 0.75H_2O$) C, H, N. IR (KBr): ν (cm^{-1}) = 3325, 1682, 1645. ES-

MS (CH_2Cl_2/MeOH/CH_3COONH_4) *m*/*z* (%): 675 (17) $[2M + H]^{+\bullet}$, 338 (100) $[MH]^{+\bullet}$.

***N*-Hydroxy-3-[2-(3,4,5-trimethoxybenzoyl)benzofuran-5-yl]-acrylamide (10c).** Yield: 0.23 g of colorless crystals (58%). Mp: 192 °C. ^{1}H NMR (DMSO-d_6): δ (ppm) = 10.80 (s, br, 1H), 9.09 (s, br, 1H), 8.04 (s, 1H), 7.91 (s, 1H), 7.85−7.76 (m, 2H), 7.61 (d, 1H, J = 15.8 Hz), 7.31 (s, 2H), 6.50 (d, 1H, J = 15.8 Hz), 3.89 (s, 6H), 3.80 (s, 3H). IR (KBr): ν (cm^{-1}) = 3444, 3309, 1676. PI-LSI-MS *m*/*z* (%): 398 (85) $[MH]^+$. HR-PI-LSI-MS calcd for $C_{21}H_{20}NO_7$ $[MH^{+\bullet}]$: 398.1140. Found 398.1230.

Preparation of 3- and 4-(2-Bromoacetyl)benzaldehyde (33a and 33b). To a solution of the respective acetylbenzaldehyde (5.00 g, 33.75 mmol, Aldrich) in CH_2Cl_2 (40 mL) a few drops of a bromine solution (33.75 mmol, 1.72 mL) in CH_2Cl_2 (10 mL) were added at room temperature. After consumption of the added bromine, the mixture was cooled to 0 °C and the remaining bromine solution was added dropwise within half an hour. The mixture was poured into water, and the organic layer was separated, washed with saturated brine (2 × 50 mL), and dried (Na_2SO_4). The solvent was removed under reduced pressure, and the product was purified by column chromatography (SiO_2, CH_2Cl_2).

4-(2-Bromoacetyl)benzaldehyde (33a). Yield: 4.60 g (60%). Mp: 154.2 °C (dec). ^{1}H NMR (DMSO-d_6): δ (ppm) = 10.13 (s, 1H), 8.20 (d, 2H, J = 8.3 Hz), 8.07 (d, 2H, J = 8.4 Hz), 5.03 (s, 2H). Anal. ($C_9H_7BrO_2$) C, H. IR (KBr): ν (cm^{-1}) = 1692, 1208. EI-MS (70 eV) *m*/*z* (%): 226 (3) $[M]^{+\bullet}$, 133 (100).

3-(2-Bromoacetyl)benzaldehyde (33b). Yield: 3.70 g (49%) of colorless crystals. Mp: 55.4−57.5 °C. ^{1}H NMR ($CDCl_3$): δ (ppm) = 10.11 (s, 1H), 8.48 (t, 1H, J = 1.6 Hz), 8.27 (dt, 1H, J = 7.7 Hz, J = 1.5 Hz), 8.14 (dt, 1H, J = 7.7 Hz, J = 1.5 Hz), 7.71 (t, 1H, 7.7 Hz). Anal. ($C_9H_7BrO_2$) C, H. IR (KBr): ν (cm^{-1}) = 1692, 1176. CI-MS (NH_3) *m*/*z* (%): 244 (59) $[M + NH_4]^+$, 133 (100).

Preparation of (Benzofuran-2-carbonyl)arylaldehydes.[40] The benzofuran-2-carbonylarylaldehydes were prepared according to the analogous literature[30] as follows: A mixture of the respective 2-bromoacetophenone (33.6 mmol), the suitable substituted 2-hydroxybenzenealdehyde (33.6 mmol), and K_2CO_3 (4.66 g, 33.7 mmol) in CH_3CN (100 mL) was refluxed for 3−5 h (TLC; SiO_2, CH_2Cl_2). The mixture was cooled to room temperature, and H_2O (200 mL) was added. The precipitating product was removed by filtration and purified by column chromatography (SiO_2, CH_2Cl_2) to yield colorless crystals.

4-(Benzofuran-2-carbonyl)benzaldehyde (34a). Yield: 1.04 g (28%). Mp: 153.5−153.6 °C. ^{1}H NMR (DMSO-d_6): δ (ppm) = 10.17 (s, 1H), 8.20−8.11 (m, 4H), 7.90−7.86 (m, 2H), 7.80 (d, 1H, J = 8.4 Hz), 7.61 (t, 1H, J = 7.8 Hz), 7.42 (t, 1H, J = 7.5 Hz). Anal. ($C_{16}H_{10}O_3$) C, H. IR (KBr): ν (cm^{-1}) = 1651, 1607. EI-MS (70 eV) *m*/*z* (%): 250 (100) $[M]^+$.

3-(Benzofuran-2-carbonyl)benzaldehyde (34b). Yield: 1.82 g (49%). Mp: 102.2−103.3 °C. ^{1}H NMR (DMSO-d_6): δ (ppm) = 10.15 (s, 1H), 8.50 (t, 1H, J = 1.5 Hz), 8.31 (d,t, 1H, J_d = 7.7 Hz, J_t = 1.5 Hz), 8.23 (d,t, 1H, J_d = 7.9 Hz, J_t = 1.5 Hz), 7.91−7.78 (m, 4H), 7.60 (t, 1H, J = 7.8 Hz), 7.42 (t, 1H, J = 7.5 Hz). Anal. ($C_{16}H_{10}NO_3$) C, H, N. IR (KBr): ν (cm^{-1}) = 1704, 1689. EI-MS (70 eV) *m*/*z* (%): 250 (100) $[M + H]^+$.

2-Benzoylbenzofuran-5-carbaldehyde (40a).[30] Yield: 7.40 g (88%) of colorless crystals. Mp: 155.0−157.2 °C. ^{1}H NMR (DMSO-d_6): δ (ppm) = 10.11 (s, 1H), 8.46 (s, 1H), 8.16−7.58 (m, 8H). Anal. ($C_{16}H_{10}O_3$) C, H. IR (pure solid): ν (cm^{-1}) = 2823, 1698, 1641. EI-MS (70 eV) *m*/*z* (%): 250 (29) $[M]^+$, 77 (100).

2-(3-Methoxybenzoyl)benzofuran-5-carbaldehyde (40b). Yield: 3.54 g (53%) of colorless crystals. Mp: 137.6 °C. ^{1}H NMR (DMSO-d_6): δ (ppm) = 10.14 (s, 1H), 8.50 (d, 1H, J = 1.3 Hz), 8.13 (dd, 1H, J = 1.6 Hz, J = 8.8 Hz), 8.05−7.95 (m, 2H), 7.69−7.50 (m, 3H), 7.39−7.31 (m, 1H), 3.90 (s, 3H). Anal. ($C_{16}H_{10}O_3$) C, H. IR (pure solid): ν (cm^{-1}) = 2841, 1698, 1656. CI-MS (NH_3) *m*/*z* (%): 281 (20) $[M + H^+]^+$, 298 (100) $[M + NH_4^+]^+$.

2-(3,4,5-Trimethoxybenzoyl)benzofuran-5-carbaldehyde (40c). Yield: 2.53 g (43%). Mp: 150−153 °C. ^{1}H NMR (DMSO-d_6): δ (ppm) = 10.12 (s, 1H), 8.48−8.45 (m, 1H), 8.13−8.06 (m, 2H), 8.00 (d, 1H, J = 8.5 Hz), 7.32 (s, 2H), 3.90 (s, 6H), 3.80 (s, 3H). Anal. ($C_{19}H_{16}O_6$) C, H. IR (pure solid): ν (cm^{-1}) = 2830, 1691, 1649. EI-MS (70 eV) *m*/*z* (%): 340 (100) $[M]^+$, 269 (20), 173 (60).

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Supporting Information Available: Additional experimental details for syntheses, analytical data, combustion analysis results, and details of biological test systems. This material is available free of charge via the Internet at http://pubs.acs.org.

References

(1) Sorbera, L. A. Epigenetic target as an approach to cancer therapy and chemoprevention. *Drugs Future* **2006**, *31*, 335−344.
(2) Yoo, C. B.; Jones, P. A. Epigenetic therapy of cancer: past, present and future. *Nat. Rev. Drug Discovery* **2006**, *5*, 37−50.
(3) Strahl, B. D.; Allis, C. D. The language of covalent histone modifications. *Nature* **2000**, *403*, 41−45.
(4) Minucci, S.; Pelicci, P. G. Histone deacetylase inhibitors and the promise of epigenetic (and more) treatments for cancer. *Nat. Rev. Cancer* **2006**, *6*, 38−51.
(5) de Ruijter, A. J. M.; van Gennip, A. H.; Caron, H. N.; Kemp, S.; van Kuilenburg, A. B. P. Histone deacetylases (HDACs): characterization of the classical HDAC family. *J. Biochem.* **2003**, *370*, 737−749.
(6) Heinzel, T.; Lavinsky, R. M.; Mullen, T. M.; Soderstrom, M.; Laherty, C. D.; Torchia, J.; Yang, W. M.; Brard, G.; Ngo, S. D.; Davie, J. R.; Seto, E.; Eisenman, R. N.; Rose, D. W.; Glass, C. K.; Rosenfeld, M. G. A complex containing N-CoR, mSin3 and histone deacetylase mediates transcriptional repression. *Nature* **1997**, *387*, 43−48.
(7) Lagger, G.; O'Carroll, D.; Rembold, M.; Khier, H.; Tischler, J.; Weitzer, G.; Schuettengruber, B.; Hauser, C.; Brunmeir, R.; Jenuwein, T.; Seiser, C. Essential function of histone deacetylase 1 in proliferation control and CDK inhibitor repression. *EMBO J.* **2002**, *21*, 2672−2681.
(8) Roeske, A.; Gekeler, V.; Beckers, T.; Stephan, C.; Jung, K.; Loening, S.; Kristiansen, G.; Dietel, M.; Denkert, C.; Weichert, W. Class I Histone Deacetylase Isoforms in Prostate Cancer: Expression Patterns in Vivo and Functional Implications in Vitro. Presented at the 18th EORTC-NCI-AACR Symposium, Prague, Czech Republic, November 7−10, 2006; Abstract 350.
(9) Johnstone, R. W.; Licht, J. D. Histone deacetylase inhibitors in cancer therapy: is transcription the primary target? *Cancer Cell* **2003**, *4*, 13−18.
(10) Sasakawa, Y.; Naoe, Y.; Sogo, N.; Inoue, T.; Sasakawa, T.; Matsuo, M.; Manda, T.; Mutoh, S. Marker genes to predict sensitivity to FK228, a histone deacetylase inhibitor. *Biochem. Pharmacol.* **2005**, *69*, 603−616.
(11) Dokmanovic, M.; Marks, P. A. Prospects: histone deacetylase inhibitors. *J. Cell. Biochem.* **2005**, *96*, 293−304.
(12) Remiszewski, S. W.; Sambucetti, L. C.; Bair, K. W.; Bontempo, J.; Cesarz, D.; Chandramouli, N.; Chen, R.; Cheung, M.; Cornell-Kennon, S.; Dean, K.; Diamantidis, G.; France, D.; Green, M. A.; Howell, K. L.; Kashi, R.; Kwon, P.; Lassota, P.; Martin, M. S.; Mou, Y.; Perez, L. B.; Sharma, S.; Smith, T.; Sorensen, E.; Taplin, F.; Trogani, N.; Versace, R.; Walker, H.; Weltchek-Engler, S.; Wood, A.; Wu, A.; Atadja, P. *N*-Hydroxy-3-phenyl-2-propenamides as novel inhibitors of human histone deacetylase with in vivo antitumor activity: discovery of (2*E*)-*N*-hydroxy-3-[4-[[(2-hydroxyethyl)[2-(1*H*-indol-3-yl)ethyl]amino]methyl]phenyl l]-2-propenamide (NVP-LAQ824). *J. Med. Chem.* **2003**, *46*, 4609−4624.
(13) Plumb, J. A.; Finn, P. W.; Williams, R. J.; Bandara, M. J.; Romero, M. R.; Watkins, C. J.; La Thangue, N. B.; Brown, R. Pharmacodynamic response and inhibition of growth of human tumor xenografts by the novel histone deacetylase inhibitor PXD101. *Mol. Cancer Ther.* **2003**, *2*, 721−728.
(14) Kelly, W. K.; O'Connor, O. A.; Krug, L. M.; Chiao, J. H.; Heaney, M.; Curley, T.; MacGregore-Cortelli, B.; Tong, W.; Secrist, J. P.; Schwartz, L.; Richardson, S.; Chu, E.; Olgac, S.; Marks, P. A.; Scher, H.; Richon, V. M. Phase I study of an oral histone deacetylase inhibitor, suberoylanilide hydroxamic acid, in patients with advanced cancer. *J. Clin. Oncol.* **2005**, *23*, 3923−3931.

(15) Buggy, J. J.; Cao, Z. A.; Bass, K. E.; Verner, E.; Balasubramanian, S.; Liu, L.; Schultz, B. E.; Young, P. R.; Dalrymple, S. A. CRA-024781: a novel synthetic inhibitor of histone deacetylase enzymes with antitumor activity in vitro and in vivo. *Mol. Cancer Ther.* **2006**, *5*, 1309−1317.
(16) Ryan, Q. C.; Headlee, D.; Acharya, M.; Sparreboom, A.; Trepel, J. B.; Ye, J.; Figg, W. D.; Hwang, K.; Chung, E. J.; Murgo, A.; Melillo, G.; Elsayed, Y.; Monga, M.; Kalnitskiy, M.; Zwiebel, J.; Sausville, E. A. Phase I and pharmacokinetic study of MS-275, a histone deacetylase inhibitor, in patients with advanced and refractory solid tumors or lymphoma. *J. Clin. Oncol.* **2005**, *23*, 3912−3922.
(17) Kalita, A.; Bonfils, C.; Maroun, C.; Fournel, M.; Rahil, G.; Yan, T. P.; Lu, A.; Reid, G. K.; Besterman, J. M.; Zuomei, L. Pharmacodynamic Assessment of MGCD0103, a Novel Isotype-Specific HDAC Inhibitor, in Preclinical Evaluations and Phase I Trials. Presented at the 2005 AACR-NCI-EORTC International Conference, Philadelphia, PA, November 14−18, 2005.
(18) Nakajima, H.; Kim, Y. B.; Terano, H.; Yoshida, M.; Horinouchi, S. FR901228, a potent antitumor antibiotic, is a novel histone deacetylase inhibitor. *Exp. Cell Res.* **1998**, *241*, 126−133.
(19) Gottlicher, M.; Minucci, S.; Zhu, P.; Kramer, O. H.; Schimpf, A.; Giavara, S.; Sleeman, J. P.; Lo Coco, F.; Nervi, C.; Pelicci, P. G.; Heinzel, T. Valproic acid defines a novel class of HDAC inhibitors inducing differentiation of transformed cells. *EMBO J.* **2001**, *20*, 6969−6978.
(20) Garber, K. HDAC inhibitors overcome first hurdle. *Nat. Biotechnol.* **2007**, *25*, 17−19.
(21) Beckers, T.; Burkhardt, C.; Wieland, H.; Gimmnich, P.; Ciossek, T.; Maier, T.; Sanders, K. Distinct pharmacological properties of 2nd generation HDAC inhibitors with the benzamide or hydroxamate head group. *Int. J. Cancer*, in press.
(22) Burgess, A.; Ruefli, A.; Beamish, H.; Warrener, R.; Saunders, N.; Johnstone, R.; Gabrielli, B. Histone deacetylase inhibitors specifically kill nonproliferating tumour cells. *Oncogene* **2004**, *23*, 6693−6701.
(23) Kramer, O. H.; Baus, D.; Knauer, S. K.; Stein, S.; Jager, E.; Stauber, R. H.; Grez, M.; Pfitzner, E.; Heinzel, T. Acetylation of Stat1 modulates NF-kappaB activity. *Genes Dev.* **2006**, *20*, 473−485.
(24) Li, Y.; Kao, G. D.; Garcia, B. A.; Shabanowitz, J.; Hunt, D. F.; Qin, J.; Phelan, C.; Lazar, M. A. A novel histone deacetylase pathway regulates mitosis by modulating Aurora B kinase activity. *Genes Dev.* **2006**, *20*, 2566−2579.
(25) Mahboobi, S.; Pongratz, H.; Hufsky, H.; Hockemeyer, J.; Frieser, M.; Lyssenko, A.; Paper, D. H.; Bürgermeister, J.; Böhmer, F.-D.; Fiebig, H.-H.; Burger, A. M.; Baasner, S.; Beckers, T. Synthetic 2-aroylindole derivatives as a new class of potent tubulin-inhibitory, antimitotic agents. *J. Med. Chem.* **2001**, *44*, 4535−4553.
(26) Zhang, J.; Xiong, C.; Ying, J.; Wang, W.; Hruby, V. J. Stereoselective synthesis of novel dipeptide β-turn mimetics targeting melanocortin peptide receptors. *Org. Lett.* **2003**, *5*, 3115−3118.
(27) Castro, B.; Dormoy, J. R.; Evin, G.; Selve, C. Peptide coupling reagents. IV. *N*-[Oxytris(dimethylamino)phosphonium]benzotriazole hexafluorophosphate. *Tetrahedron Lett.* **1975**, *14*, 1219−1222.
(28) Kamitori, Y.; Hojo, M.; Masuda, R.; Yoshida, T. Alumina as an versatile catalyst for the selective acetalization of aldehydes. *Tetrahedron Lett.* **1985**, *26*, 4767−4770.
(29) Maya, A. B. S.; Perez-Melero, C.; Mateo, C.; Alonso, D.; Fernandez, J. L.; Gajate, C.; Mollinedo, F.; Pelaez, R.; Caballero, E.; Medarde, M. Further naphthylcombretastatins. An investigation on the role of the naphthalene moiety. *J. Med. Chem.* **2005**, *48*, 556−568.
(30) Stille, J. R.; Ward, J. A.; Leffelman, C.; Sullivan, K. A. 5-Formylsalicylaldehyde as a linker for the synthesis of benzofuran containing insulin senstivity enhancer compounds. *Tetrahedron Lett.* **1996**, *37*, 9267−9270.
(31) Somoza, J. R.; Skene, R. J.; Katz, B. A.; Mol, C.; Ho, J. D.; Jennings, A. J.; Luong, C.; Arvai, A.; Buggy, J. J.; Chi, E.; Tang, J.; Sang, B. C.; Verner, E.; Wynands, R.; Leahy, E. M.; Dougan, D. R.; Snell, G.; Navre, M.; Knuth, M. W.; Swanson, R. V.; McRee, D. E.; Tari, L. W. Structural snapshots of human HDAC8 provide insights into the class I histone deacetylases. *Structure* **2004**, *12*, 1325−1334.
(32) Schmidt, M.; Lu, Y.; Liu, B.; Fang, M.; Mendelsohn, J.; Fan, Z. Differential modulation of paclitaxel-mediated apoptosis by p21waf1 and p27kip1. *Oncogene* **2000**, *19*, 2423−2429.
(33) Dignam, J. D.; Lebovitz, R. M.; Roeder, R. G. Accurate transcription initiation by RNA polymerase II in a soluble extract from isolated mammalian nuclei. *Nucleic Acids Res.* **1983**, *11*, 1475−1489.
(34) Wegener, D.; Wirsching, F.; Riester, D.; Schwienhorst, A. A fluorogenic histone deacetylase assay well suited for high-throughput activity screening. *Chem. Biol.* **2003**, *10*, 61−68.
(35) Braunger, J.; Zoche, M.; Schmidt, M.; Wiesbacher, M.; Burkhardt, C.; Gekeler, V.; Beckers, T. High Content Profiling of HDAC Inhibitors in Cellular Assays. Presented at the 94th Annual Meeting of the American Association for Cancer Research, Washington, DC, July 11−14, 2003.
(36) O'Brian, J.; Wilson, I.; Orton, T.; Pognan, F. Investigation of the Alamar Blue (resazurin) fluorescent dye for the assessment of mammalian cell toxicity. *Eur. J. Biochem.* **2000**, *267*, 5421−5426.
(37) Kyhse-Anderson, J. Electroblotting of multiple gels: a simple apparatus without buffer tank for rapid transfer of proteins from polyacrylamide to nitrocellulose. *J. Biochem. Biophys. Methods* **1984**, *10*, 203−209.
(38) Mahboobi, S.; Teller, S.; Pongratz, H.; Hufsky, H.; Sellmer, A.; Botzki, A.; Uecker, A.; Beckers, T.; Baasner, S.; Schachtele, C.; Ueberall, F.; Kassack, M. U.; Dove, S.; Böhmer, F.-D. Bis(1*H*-2-indolyl)methanones as a novel class of inhibitors of the platelet-derived growth factor receptor kinase. *J. Med. Chem.* **2002**, *45*, 1002−1018.
(39) Moyer, M. P.; Shiurba, J. F.; Rapoport, H. Metal−halogen exchange of bromoindoles. A route to mono substituted Indoles. *J. Org. Chem.* **1986**, *51*, 5106−5110.
(40) Sun, L.; Vasilevich, N. I.; Fuselier, J. A.; Coy, D. H. Abilities of 3,4-diarylfuran-2-one analogs of combretastatin A-4 to inhibit both proliferation of tumor cell lines and growth of relevant tumors in nude mice. *Anticancer Res.* **2004**, *24*, 179−186.

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