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Total Synthesis of Resorcinol Amide Hsp90 Inhibitor AT13387

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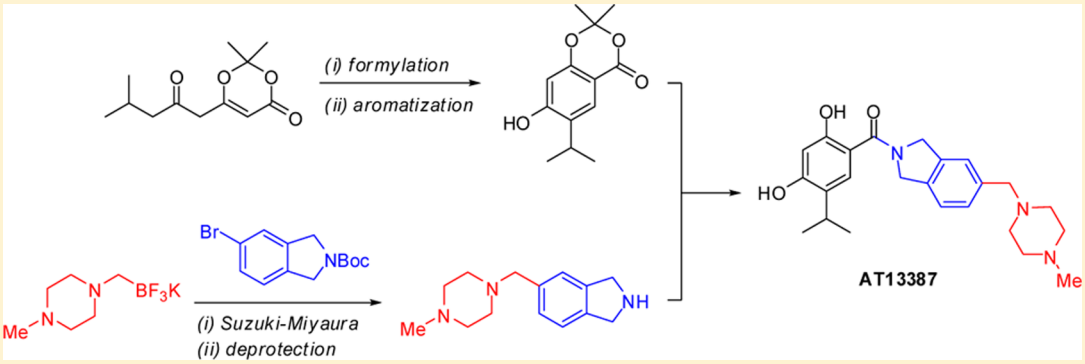
Supporting Information



ABSTRACT: The synthesis of *C*-5-substituted resorcinol amide AT13387, a known Hsp90 inhibitor currently in clinical trials, is reported without the use of phenolic protection in an overall yield of 13.4%. Biomimetic aromatization and Suzuki-Miyaura cross coupling approach were employed to synthesize the resorcinol and *iso*-indoline units, respectively, which were efficiently coupled using Grignard-mediated amidation.

INTRODUCTION

Molecular chaperones are responsible for the correct folding, stabilization and function of other cellular proteins.[1] The most extensively studied of these is heat shock protein 90 (Hsp90), which is a highly abundant 90-kDa protein and regulates the conformation, activation, function and stability of client proteins.[2] Over recent years, Hsp90 has gained considerable interest due to the recognition of its importance to cancer cell survival.[3] Over the past decade a substantial number of Hsp90 inhibitors have been discovered with one of the earliest being the resorcylate natural product radicicol (**1**) (Figure 1).[2] The

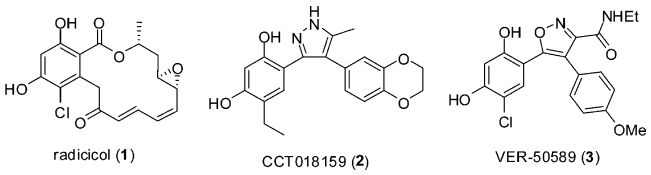


Figure 1. Structures of resorcinol-containing Hsp90 inhibitors.

resorcinol-diarylpyrazole CCT018159 (**2**) was identified as a Hsp90 inhibitor through HTS, and computer-aided design resulted in the discovery of the more potent resorcinylic isoxazole amide VER-50589 (**3**). Moreover, further semi-synthetic structure activity relationship (SAR) studies have been reported on radicicol (**1**) and its analogues.[4]

It became evident that the resorcinol anchor unit was critical for binding, especially in the effectiveness of radicicol as an Hsp90 inhibitor.[2] Several pharmaceutical companies, including Pfizer[5] and Astex Therapeutics[6] undertook medicinal chemistry research programs on Hsp90. Their efforts have led to lead structures **4** and **5**, differing by the *C*-5 resorcinol substituent (Figure 2). The study by Pfizer scientists led to the

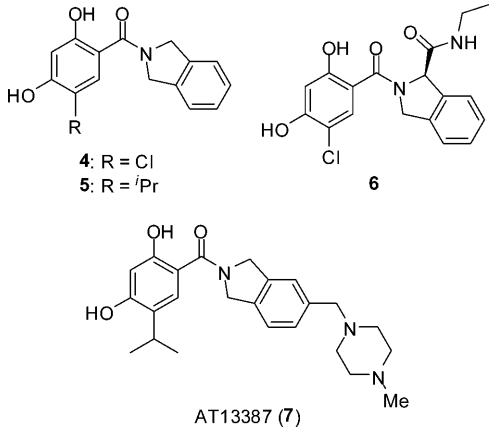


Figure 2. Structures of Pfizer and Astex Hsp90 inhibitors.

identification of diamide **6**, which was potent (K_i < 1 nM, cell IC_{50} = 0.3 μM) and also displayed good clearance and acceptable oral bioavailability.[7] Astex used fragment-based drug discovery (FBDD) and focused SAR studies of their lead resorcylate **5** to develop AT13387 (**7**) (K_d = 0.71 nM, HCT116 cell IC_{50} = 48 nM, hERG % inhibition: 7% at 3 μM),[8] which

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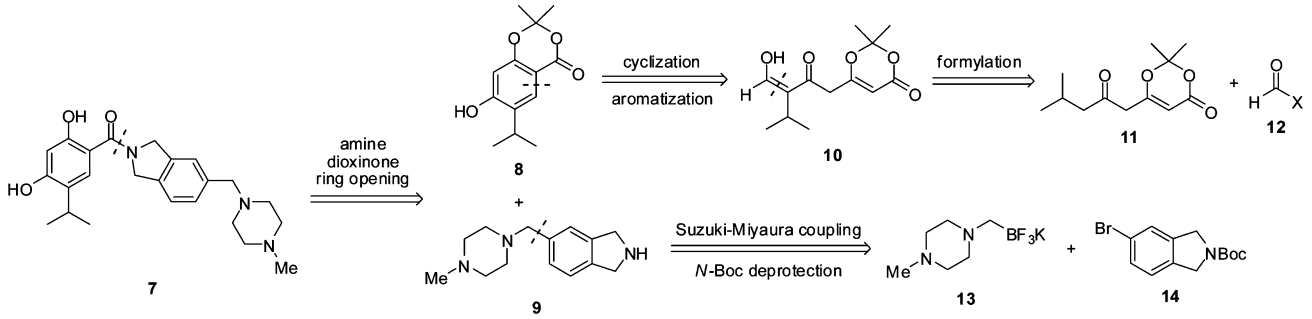
Scheme 1. Projected Retrosynthetic Strategy



has progressed through preclinical development, and is now in clinical trials for the treatment of refractory gastrointestinal stomal tumors.

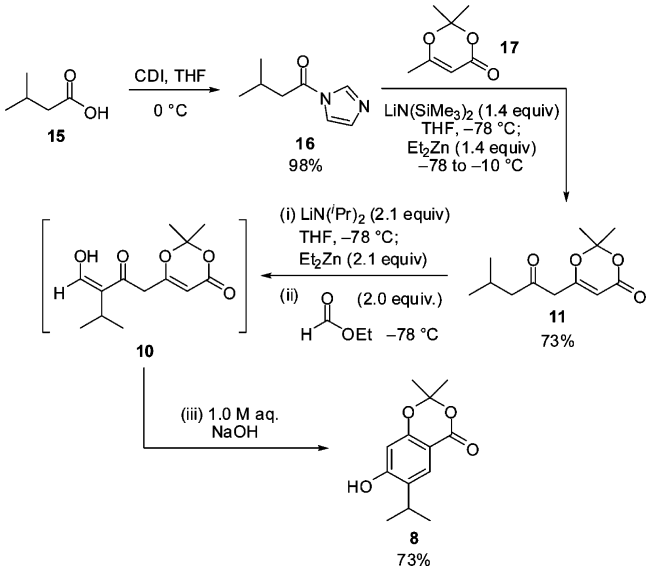
The resorcylate unit of these molecules has previously been synthesized from substituted benzoic acid precursors with lengthy sequences of transformations including aromatic substitution, palladium catalyzed coupling reactions and protection−deprotection steps. For example, the reported total synthesis of AT13387 is 13 steps with an overall yield of 2.6%.[9] We sought to circumvent these issues by the use of our biomimetic approach[10] to generate the resorcylate ring thereby developing a flexible synthesis much more suitable for analogue generation as well as for the scale up synthesis of lead compounds. We considered that AT13387 (**7**) should be available from dioxinone-resorcylate **8** and amine **9** via transacylative ring-opening (Scheme 1). The *C*-5 *iso*-propyl dioxinone-resorcylate **8** should in turn be prepared from dioxinone **10** utilizing a base-mediated cyclization-aromatization process. Intermediate **10** should be accessible from the *C*-formylation of the dilithium enolate of **11** with esters, activated amides or mixed anhydrides **12**. Finally, *iso*-indoline **9** should be available using Molander's methodology,[11] from potassium 4-methylpiperazinomethyltrifluoroborate (**13**) and bromo-*iso*-indoline **14**.

RESULTS AND DISCUSSION

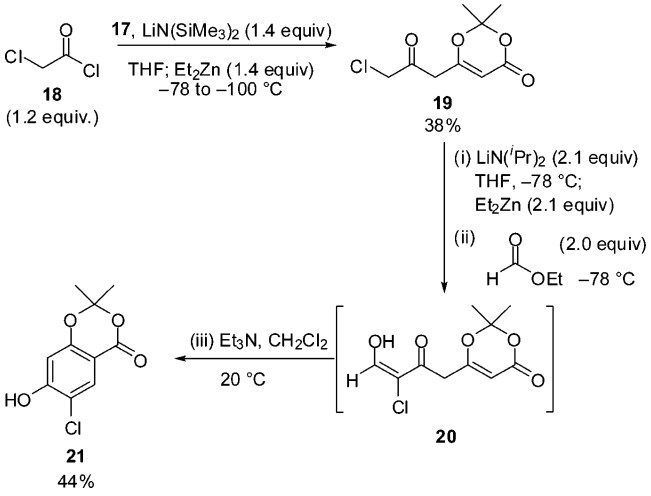
iso-Valeric acid (**15**) was converted to the imidazolide **16** using carbonyl diimidazole (98% yield), which was allowed to react with the lithium enolate, generated from dioxinone **17** with lithium bis-trimethylsilylamide in the presence of diethylzinc, to give keto-dioxinone **11** in 73% yield on a multigram scale (Scheme 2).[12] The zinc enolate dianion, derived from keto-dioxinone **11** with lithium di-*iso*-propylamide and diethylzinc, was allowed to react with ethyl formate (2 equiv) to give crude adduct **10**, which was directly aromatized with aqueous sodium hydroxide followed by acidification with hydrochloric acid to give dioxinone-resorcylate **8** (73% yield), without the need for chromatographic purification.[13]

We sought to extend this resorcylate synthesis to the equivalent Pfizer intermediate **21**. Chloroacetyl chloride (**18**) was allowed to react with the lithium enolate from dioxinone **17** to give chloro-dioxinone **19**,[14] with the best albeit modest yield (38%) being obtained with addition of chloroacetyl chloride at −100 °C (Scheme 3).[15] Chloro-dioxinone **19** was subsequently subjected to the formylation-cyclization-aromatization sequence, which gave rise to enol **20**. Without isolation, final reaction with triethylamine gave the dioxinone-resorcylate **21** in 44% yield over the two steps.[16]

Scheme 2. Synthesis of Key Resorcylate 8 using a Formylation−Cyclization−Aromatization Sequence



Scheme 3. Synthesis of Key Resorcylate 21 from Chloroacetyl Chloride



At this stage, we examined the synthesis of the *iso*-indoline unit (Scheme 4). Commercially available 5-bromophthalimide (**22**) was allowed to react with sodium borohydride and boron trifluoride etherate in THF to give 5-bromo-*iso*-indoline **23** (76%), which was subsequently protected as the corresponding

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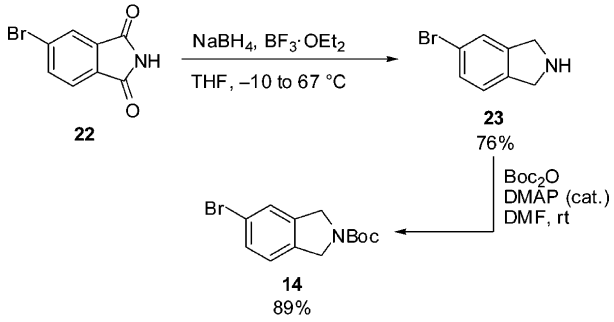
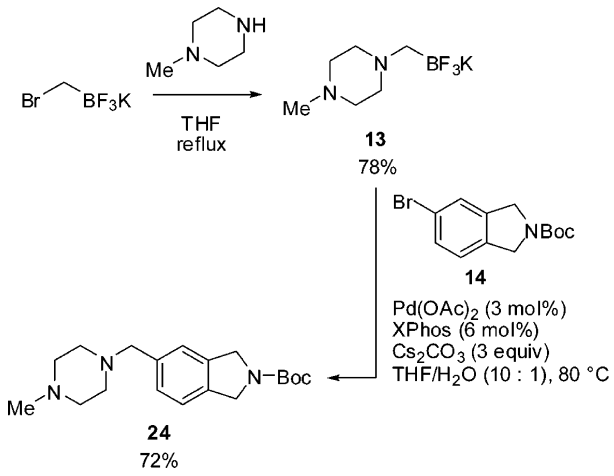
Scheme 4. Synthesis of *N*-Boc Protected 5-Bromo-*iso*-indoline 14



Boc-derivative **14** (89%) with di-*tert*-butyl dicarbonate in DMF.[17]

Following the excellent Molander protocol, potassium bromomethyltrifluoroborate was allowed to react with *N*-methylpiperazine under reflux in THF to provide potassium 4-methylpiperazinomethyl-trifluoroborate (**13**) in 78% yield (Scheme 5). Much to our delight, subsequent Suzuki-Miyaura

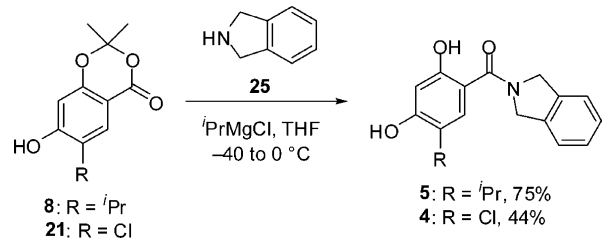
Scheme 5. Synthesis of Piperazino-*iso*-indoline 24 using a Suzuki-Miyaura Coupling Reaction



cross coupling with *N*-Boc-5-bromo-*iso*-indoline **14** using palladium acetate, dicyclohexyl-2-(2,4,6-tri*iso*-propylphenyl)-phenylphosphine (XPhos) and cesium carbonate gave the piperazino-*iso*-indoline **24** in 72% yield.[11]

Iso-indoline **25** was initially used as a model amine to investigate the resorcinol amide formation from dioxinone-resorcylates **8** and **21** (Scheme 6). To our delight, addition of *iso*-propylmagnesium chloride (2.2 equiv) to a mixture of

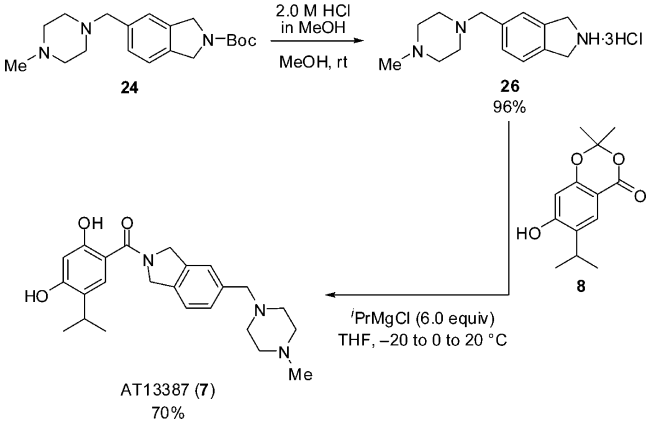
Scheme 6. Synthesis of Hsp90 Inhibitor Lead Resorcylamides 5 and 4



resorcylate **8** and amine **25** at −40 °C and warming up to 0 °C, gave amide **5** (75%). Similarly, chlororesorcylate **4** was obtained, albeit in lower yield (44%), presumably due to side reactions of the Grignard reagent with the aryl chloride.

The *N*-Boc protected 5-*N*-methylpiperazine *iso*-indoline **24** was allowed to react with methanolic HCl to afford trihydrochloride salt **26** in 96% yield.[18] Subsequently, salt **26** was allowed to react with dioxinone-resorcylate **8** and *iso*-propylmagnesium chloride (6.0 equiv) at −20 °C, gradually warming up to 0 °C and subsequently at room temperature, to give AT13387 (**7**) in 70% yield (Scheme 7).

Scheme 7. Completion of the Synthesis of Hsp90 Inhibitor AT13387 (7)



In conclusion, we have completed a total synthesis of AT13387 (**7**), with 9 synthetic manipulations and an overall yield of 13% using our biomimetic aromatization, a key Suzuki-Miyaura palladium cross coupling for the *iso*-indoline unit, and an *iso*-propylmagnesium chloride mediated amine dioxinone transacylation reaction. Further studies highlighting the applicability of this methodology to other bioactive resorcylates will be reported in due course.

EXPERIMENTAL SECTION

General Methods. All reactions were carried out in oven-dried glassware under N_2, using commercially supplied solvents and reagents unless otherwise stated. Reaction temperatures other than room temperature were recorded as the external bath temperature unless otherwise stated. THF, CH_2Cl_2, Et_3N, and MeOH were redistilled from Na-Ph_2CO, CaH_2, CaH_2, and Mg turnings−I_2, respectively. Petroleum spirits bp 40−60 °C was used. Column chromatography was carried out on silica gel, using flash techniques (eluants are given in parentheses). Analytical thin layer chromatography was performed on precoated silica gel F_{254} aluminum plates with visualization under UV light or by staining using acidic vanillin, anisaldehyde, potassium permanganate or ninhydrin spray reagents. Mps were obtained using a melting point apparatus and are uncorrected. Infrared data were carried out neat unless otherwise stated. Indicative features of each spectrum are given with adsorptions reported in wavenumbers (cm^{-1}). ^1H NMR spectra were recorded at 400 or 500 MHz with chemical shifts (δ) quoted in parts per million (ppm) and coupling constants (J) recorded in Hertz (Hz). ^{13}C NMR spectra were recorded at 101 or 126 MHz with chemical shifts (δ) quoted in ppm. High resolution mass spectra (electrospray ionization, ESI-TOF) were recorded by Imperial College Mass Spectrometry Service.

*1-(3-Methylbutanoyl)-imidazole (**16**).* Carbonyl diimidazole (90%; 9.1 g, 50.6 mmol) was added portionwise with stirring over 15 min to *iso*-valeric acid (5.08 mL, 46 mmol) in THF (90 mL) at 0 °C. After 2 h, the mixture was diluted with Et_2O (250 mL), H_2O (100 mL) was

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added carefully and the layers separated. The organic layer was washed with H_2O (2 × 80 mL) and brine (100 mL), dried ($MgSO_4$) and rotary evaporated to give the imidazolide **16** (6.82 g, 98%) as a colorless oil: R_f 0.15 (petroleum spirits/EtOAc 1:1); ^{1}H NMR (acetone-d_6, 400 MHz) δ 8.30 (s, 1H), 7.64 (br m, 1H), 7.03 (br m, 1H), 2.94 (d, J = 6.8 Hz, 2H), 2.26 (m, 1H), 1.03 (d, J = 6.4 Hz, 6H); ^{13}C NMR (acetone-d_6, 101 MHz) δ 171.3, 138.4, 132.3, 118.0, 45.0, 26.9, 23.5 (2C);[19] used in the next step without further purification.

*2,2-Dimethyl-6-(4-methyl-2-oxopentyl)-4H-1,3-dioxin-4-one (**11**).* *n*-BuLi in hexanes (2.5 M; 20.5 mL, 51.3 mmol) was added dropwise with stirring to $HN(SiMe_3)_2$ (10.8 mL, 51.3 mmol) in THF (105 mL) at −78 °C. After 20 min and a further 1 h, respectively, dioxinone **17** (5.21 g, 36.7 mmol) in THF (12 mL) and Et_2Zn in hexanes (1.0 M; 51.3 mL, 51.3 mmol) were added slowly, and after a further 20 min, the mixture was allowed to warm up to −20 °C. Imidazolide **16** (6.69 g, 44 mmol) in THF (50 mL) was added and the reaction mixture immediately warmed to −10 °C. After 3.5 h, the reaction was quenched with 1.0 M aqueous HCl (100 mL), and the aqueous layer acidified to pH 1−2 using 1.0 M aqueous HCl. The product was extracted with EtOAc (2 × 100 mL) and the combined organic extracts dried ($MgSO_4$), the solvent evaporated under vacuum and the residue chromatographed (petroleum spirits/Et_2O 6:1 to 4:1 to 1:1) to give *iso*-butyl-keto-dioxinone **11** (6.07 g, 73%) as a yellow oil: R_f 0.55 (petroleum spirits:Et_2O 1:1); IR 1719, 1635 cm^{-1}; ^{1}H NMR (acetone-d_6, 400 MHz) δ 5.39 (s, 1H), 3.54 (s, 2H), 2.48 (d, J = 6.8 Hz, 2H), 2.14 (m, 1H), 1.69 (s, 6H), 0.93 (d, J = 6.8 Hz, 6H); ^{13}C NMR (acetone-d_6, 101 MHz) δ 204.8, 167.3, 161.6, 108.4, 98.1, 53.1, 48.7, 26.1 (2C), 25.9, 23.6 (2C); HRMS (ESI-TOF) m/z: $[M + H]^+$ Calcd for $C_{12}H_{18}O_4$ 227.1283; Found 227.1276.[20]

*6-(3-Chloro-2-oxopropyl)-2,2-dimethyl-4H-1,3-dioxin-4-one (**19**).* *n*-BuLi in hexanes (2.5 M; 5.6 mL, 14.0 mmol) was added dropwise with stirring to $(Me_3Si)_2NH$ (2.95 mL, 14.0 mmol) in THF (35 mL) at −78 °C. After 20 min and a further 1 h respectively, dioxinone **17** (1.42 g, 10.0 mmol) in THF (5 mL) and Et_2Zn in hexanes (1.0 M; 14.0 mL, 14.0 mmol) were added dropwise, and after a further 20 min, the mixture was cooled down to −100 °C. Chloroacetyl chloride (**18**) (0.96 mL, 12.0 mmol) was added dropwise and the reaction mixture stirred at −100 °C. After 2 h, the reaction was quenched with 1.0 M aqueous HCl (20 mL), and the aqueous layer acidified to pH 1−2 using 1.0 M aqueous HCl. The product was extracted with EtOAc (2 × 50 mL) and the combined organic extracts dried ($MgSO_4$), the solvent evaporated under vacuum and the residue chromatographed (petroleum spirits/Et_2O 4:1 to 2:1 to 1:1) to give chloro-keto-dioxinone **19** (0.82 g, 38%) as a yellow gum: R_f 0.32 (petroleum spirits:Et_2O 1:1); IR 1721, 1638 cm^{-1}; ^{1}H NMR (acetone-d_6, 400 MHz) δ 5.42 (s, 1H), 4.53 (s, 2H), 3.74 (s, 2H), 1.68 (s, 6H); HRMS (ESI-TOF) m/z: $[M + NH_4]^+$ Calcd for $C_9H_{11}ClO_4$ 236.0690; Found 236.0696.[14]

*5-Bromo-iso-indoline (**23**).* Sodium borohydride (3.48 g, 92.0 mmol) was added with stirring to 5-bromopthalamide **22** (2.0 g, 8.8 mmol) in THF (87 mL) and the resultant suspension cooled to −10 °C. $BF_3.Et_2O$ (12.7 mL, 102.6 mmol) was added slowly and once the addition was completed, the reaction mixture was heated at 70 °C. After 16 h, the reaction mixture was allowed to cool to 0 °C, quenched slowly with cold water (18 mL), diluted with EtOAc (140 mL) and made alkaline to pH 10 using 6.0 M aqueous NaOH. The organic layer was separated, washed with brine (4 × 70 mL), dried (Na_2SO_4) and rotary evaporated. The residual gray oil was diluted with Et_2O (50 mL) and acidified to pH 2 using 6.0 M aqueous HCl with stirring. The aqueous layer was separated, made alkaline to pH 10 using 6.0 M aqueous NaOH and extracted with EtOAc (70 mL). The organic layer was separated, washed with brine (3 × 70 mL), dried (Na_2SO_4) and rotary evaporated to give the 5-bromo-*iso*-indoline **23** (1.33g, 76%) as a pale green oil: R_f 0.10 (petroleum spirits/EtOAc 1:1); IR 1604 cm^{-1}; ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.38 (s, 1H), 7.32 (d, J = 7.6 Hz, 1H), 7.11 (d, J = 8.0 Hz, 1H), 4.20 (s, 2H), 4.17 (s, 2H), 2.11 (br s, 1H); ^{13}C NMR ($CDCl_3$, 101 MHz) δ 144.3, 140.9, 129.6, 125.5, 123.7, 120.4, 52.8, 52.6; HRMS (ESI-TOF) m/z: $[M(^{79}Br) + H]^+$ Calcd for $C_8H_8{}^{79}BrN$ 197.9918; Found 197.9926; m/z: $[M(^{81}Br) + H]^+$ Calcd for $C_8H_8{}^{81}BrN$ 199.9898; Found 199.9908.[17]

*tert-Butyl 5-Bromo-iso-indoline-2-carboxylate (**14**).* $(Boc)_2O$ (1.1 g, 5.04 mmol) was added to *iso*-indoline **23** (0.83 g, 4.2 mmol) in DMF (11.8 mL) at room temperature, followed by a few crystals of DMAP. After 16 h, the reaction mixture was diluted with EtOAc (50 mL), washed with brine (4 × 30 mL), dried (Na_2SO_4), the solvent evaporated under vacuum and the residue chromatographed (petroleum spirits/Et_2O 4:1 to 3:1) to give the *N*-Boc-protected product **14** (1.05 g, 84%) as a pale yellow solid: mp 54−56 °C (petroleum spirits/Et_2O 3:1); R_f 0.80 (petroleum spirits/Et_2O 1:1); IR 1697 cm^{-1}; ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.37 (m, 2H), 7.13 (d, J = 8.0 Hz, 0.5H, rotamer 1), 7.08 (d, J = 8.0 Hz, 0.5H, rotamer 2), 4.61 (m, 4H), 1.50 (s, 9H); ^{13}C NMR ($CDCl_3$, 101 MHz) δ (mixture of rotamers) 154.3, 139.6, 139.2, 136.3, 136.0, 130.4 (2C), 126.0, 125.7, 124.3, 124.0, 121.1, 79.9, 51.9, 51.6, 28.5 (3C) HRMS (ESI-TOF) m/z: $[M(^{79}Br) - {}^tBu + H]^+$ Calcd for $C_{13}H_{16}{}^{79}BrNO_2$ 241.9817; Found 241.9820; $[M(^{81}Br) - {}^tBu + H]^+$ Calcd for $C_{13}H_{16}{}^{81}BrNO_2$ 243.9801; Found 243.9796; $[M(^{79}Br) - {}^tBu + MeCN + H]^+$ Calcd for $C_{13}H_{16}{}^{79}BrNO_2$ 283.0082; Found 283.0081; $[M(^{81}Br) - {}^tBu + MeCN + H]^+$ Calcd for $C_{13}H_{16}{}^{81}BrNO_2$ 285.0062; Found 283.0062.[17]

*Potassium 1-Methyl-4-trifluoroboratomethylpiperazine (**13**).* K-(F_3BCH_2Br) (1.0 g, 5.00 mmol) was added to a solution of 1-methylpiperazine (0.59 g, 5.25 mmol) in THF (7 mL) at room temperature. After 3.5 h, the resulting mixture was the solvent evaporated under vacuum and the residue dissolved in a solution of dry acetone (150 mL) and K_2CO_3 (0.69 g, 5.0 mmol) and stirred for 30 min. The solution was filtered through a pad of Celite to remove the insoluble salts, and the filtrate was rotary evaporated. The solid product was dried under vacuum overnight to give the desired compound **13** (0.76 g, 70%) as a yellow solid: mp 117−120 °C; R_f 0.05 (petroleum spirits/EtOAc 1:1); IR 1457 cm^{-1}; ^{1}H NMR (acetone-d_6, 400 MHz) δ 3.58 (br m, 2H), 3.07 (br m, 2H), 2.91 (br m, 2H), 2.47 (br s, 2H), 2.29 (s, 3H), 2.13 (br s, 2H); HRMS (ESI-TOF) m/z: $[M - K]^-$ Calcd for $C_6H_{13}BF_3KN_2$ 181.1124; Found 181.1121.[11]

*tert-Butyl 5-((4-Methylpiperazin-1-yl)methyl)-iso-indoline-2-carboxylate (**24**).* $Pd(OAc)_2$ (5 mg, 0.022 mmol), 2-(2,4,6-*iso*-$Pr_3C_6H_2$)-P(*c*-hexyl)$_2$ (XPhos) (21 mg, 0.045 mmol), the piperazine derivative **13** (0.16 g, 0.74 mmol), and Cs_2CO_3 (0.72 g, 2.2 mmol). The tube was sealed with a septum and purged with N_2. A solution of compound **14** (0.22 g, 0.74 mmol) in THF and H_2O (10:1) (0.25 M, 3.0 mL) was added and the mixture was stirred at 80 °C. After 24 h, the reaction mixture was cooled to room temperature, diluted with H_2O (3 mL) and extracted with EtOAc (2 × 15 mL). The organic layer was dried (Na_2SO_4), the solvent evaporated under vacuum and the residue chromatographed (petroleum spirits/EtOAc:Et_3N 2:1:0.1) to yield the carbamate **24** (177 mg, 72%) as a pale yellow gum: R_f 0.15 (petroleum spirits/EtOAc:Et_3N 2:1:0.1); IR 1697 cm^{-1}; ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.24−7.14 (m, 3H), 4.67 (s, 2H), 4.62 (s, 2H), 3.50 (s, 2H), 2.80−2.28 (br s, 8H), 2.18 (s, 3H), 1.51 (s, 9H); ^{13}C NMR ($CDCl_3$, 101 MHz) δ (mixture of rotamers) 154.6, 137.7, 137.6, 137.5, 137.1, 136.2, 135.8, 128.4, 123.5, 123.2, 122.5, 122.2, 79.6, 62.9, 55.1 (2C), 53.1 (2C), 52.2, 52.1, 51.9, 51.8, 46.0, 28.5 (3C); HRMS (ESI-TOF) m/z: $[M + H]^+$ Calcd for $C_{19}H_{29}N_3O_2$ 332.2338; Found 332.2334.

*5-((4-Methylpiperazin-1-yl)methyl)iso-indoline trihydrochloride (**26**).* HCl in MeOH (2.0 M; 1.95 mL, 3.90 mmol) was added to carbamate **24** (0.13 g, 0.39 mmol) in MeOH (5.85 mL) at room temperature. After 16 h, the solvent was evaporated to give trihydrochloride salt **26** (0.13 mg, 96%) as an off-white solid: mp > 300 °C; IR 1402, 1365 cm^{-1}; ^{1}H NMR (DMSO-d_6, 400 MHz) δ 12.89−10.91 (br m, 2H), 10.03 (br m, 2H), 7.60 (br m, 2H), 7.47−7.46 (br d, J = 6.0 Hz, 1H), 4.51 (br s, 4H), 4.42−4.03 (br s, 2H), 3.47 (s, 8H), 2.78 (s, 3H); ^{13}C NMR (DMSO-d_6, 101 MHz) δ 135.5, 131.6, 131.0, 125.5, 125.5, 123.2, 58.6, 49.7 (2C), 49.7 (2C), 48.0, 47.9, 42.1; HRMS (ESI-TOF) m/z: $[M - 3HCl + H]^+$ Calcd for $C_{14}H_{24}Cl_3N_3$ 232.1814; Found 232.1804; Anal. Calcd. for $C_{14}H_{24}Cl_3N_3$: C, 49.35; H, 7.10; N, 12.33. Found: C, 49.22; H, 6.98; N, 12.24.

*7-Hydroxy-6-iso-propyl-2,2-dimethyl-4H-benzo[d][1,3]dioxin-4-one (**8**).* *n*-BuLi in hexanes (2.5 M; 4.4 mL, 11 mmol) was added dropwise with stirring to *iso*-Pr_2NH (1.55 mL, 11 mmol) in THF (40 mL) at −78 °C. After 20 min and a further 50 min respectively, keto-

dioxinone **11** (1.13 g, 5.0 mmol) in THF (10 mL) and Et_2Zn in hexanes (1.0 M; 11 mL, 11 mmol) were added dropwise with stirring at −78 °C. After 30 min, ethyl formate (0.81 mL, 10 mmol) was added slowly and the reaction mixture stirred for 2 h at −78 °C. The reaction was quenched with saturated 1.0 M aqueous NaOH (35 mL), and the reaction mixture allowed to warm up to room temperature and stirred for 10 min. The aqueous layer was acidified to pH 1−2 using 1.0 M aqueous HCl. The product was extracted with EtOAc (300 mL), washed with brine (100 mL), dried ($MgSO_4$), the solvent evaporated under vacuum and the residue triturated with Et_2O/petroleum spirits (1:9; 20 mL) and then CH_2Cl_2/petroleum spirits (1:9; 20 mL) to give dioxinone-resorcylate **8** (0.86 g, 73%) as a pale yellow solid: mp 136−138 °C (petroleum spirits/Et_2O 2:1); R_f 0.52 (petroleum spirits/Et_2O 1:1); IR 3361, 1708, 1611, 1505 cm^{-1}; ^1H NMR (acetone-d_6, 400 MHz) δ 9.61 (br s, 1H), 7.65 (s, 1H), 6.46 (s, 1H), 3.24 (sep, J = 6.8 Hz, 1H), 1.66 (s, 6H), 1.23 (d, J = 6.8 Hz, 6H); ^{13}C NMR (acetone-d_6, 101 MHz) δ 163.8, 162.0, 157.7, 132.7, 128.8, 107.6, 107.1, 104.1, 28.3, 26.8 (2C), 23.7 (2C); HRMS (ESI-TOF) m/z: $[M + H]^+$ Calcd for $C_{13}H_{16}O_4$: 237.1127; Found 237.1116; Anal. Calcd. for $C_{13}H_{16}O_4$: C, 66.09; H, 6.83. Found: C, 66.16; H, 6.87.

*6-Chloro-7-hydroxy-2,2-dimethyl-4H-benzo[d][1,3]dioxin-4-one (**21**).* *n*-BuLi in hexanes (2.5 M; 0.88 mL, 2.2 mmol) was added dropwise with stirring to *iso*-Pr_2NH (0.31 mL, 2.2 mmol) in THF (7 mL) at −78 °C. After 20 min and a further 50 min respectively, keto-dioxinone **19** (0.22 g, 1.0 mmol) in THF (1.5 mL) and Et_2Zn in hexanes (1.0 M; 2.2 mL, 2.2 mmol) were added dropwise with stirring at −78 °C. After 30 min, EtOCHO (0.16 mL, 2.0 mmol) was added slowly and the mixture stirred for 2 h at −78 °C. The reaction was quenched with saturated 1.0 M aqueous HCl (25 mL), and the aqueous layer acidified to pH 1−2 using 1.0 M aqueous HCl. The product was extracted with EtOAc (50 mL), washed with brine (20 mL), dried ($MgSO_4$) and rotary evaporated to give the crude formyl-keto-dioxinone **20**, which was dissolved in CH_2Cl_2 (22 mL), Et_3N (2 mL) added, and the mixture stirred at room temperature. After 16 h, 1.0 M aqueous HCl (40 mL) and EtOAc (75 mL) were added and the layers separated. The aqueous layer was further extracted with EtOAc (25 mL) and the combined organic extracts dried ($MgSO_4$), the solvent evaporated under vacuum and the residue chromatographed (petroleum spirits/EtOAc 5:1 to 1:1) to give dioxinone-resorcylate **21** (100 mg, 44%) as a pale yellow solid; mp 138−140 °C (petroleum spirits/Et_2O 2:1); R_f 0.40 (petroleum spirits/Et_2O 1:1); IR 3300, 1684, 1604 cm^{-1}; ^1H NMR (acetone-d_6, 400 MHz) δ 10.19 (br s, 1H), 7.82 (s, 1H), 6.66 (s, 1H), 1.71 (s, 6H); ^{13}C NMR (acetone-d_6, 101 MHz) δ 161.8, 160.9, 158.2, 132.1, 117.5, 108.4, 106.0 (2C), 26.8 (2C); HRMS (ESI-TOF) m/z: $[M + H]^+$ Calcd for $C_{10}H_8ClO_4$ 228.0189; Found 228.0182.

General Procedure for the Amine Dioxinone Ring Opening. The dioxinone-resorcylate (0.25 mmol) in THF (1.5 mL) was cooled to −40 °C, when amine **25** (0.25 mmol) in THF (0.2 mL), and after 10 min, ^{i}PrMgCl in THF (2.0 M; 0.28 mL, 0.55 mmol) were added dropwise. The mixture was warmed up to −10 °C, and after 1 h, the temperature was increased to 0 °C. After 0.5−1.5 h, the reaction mixture was quenched with saturated aqueous NH_4Cl (5 mL), the aqueous layer acidified to pH 1−2 using aqueous HCl (1.0 M) and extracted with EtOAc (25 mL). The organic layer was washed with brine (20 mL), dried ($MgSO_4$), the solvent evaporated under vacuum and the residue triturated with Et_2O (25 mL) to give the desired resorcylamide.

*(2,4-Dihydroxy-5-iso-propylbenzoyl)-iso-indoline (**5**).* Yield 75%; white solid; mp 104−106 °C (petroleum spirits/Et_2O 2:1); R_f 0.48 (petroleum spirits/Et_2O 1:1); IR 3255, 1627, 1562 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz) δ 10.03 (s, 1H), 9.60 (s, 1H), 7.33 (br m, 4H), 7.04 (s, 1H), 6.40 (s, 1H), 4.77 (br s, 4H), 3.09 (sep, J = 6.8 Hz, 1H), 1.13 (d, J = 6.8 Hz, 6H); ^{13}C NMR (DMSO-d_6, 101 MHz) δ 168.7, 156.7, 153.7, 136.8, 136.0, 127.2 (2C), 125.5, 125.3, 122.7 (2C), 113.9, 102.4, 52.9, 51.7, 25.8, 22.6 (2C); HRMS (ESI-TOF) m/z: $[M + H]^+$ Calcd for $C_{18}H_{19}NO_3$ 298.1443; Found 298.1438; Anal. Calcd. for $C_{18}H_{19}NO_3$: C, 72.71; H, 6.44; N, 4.71. Found: C, 72.82; H, 6.38; N, 4.81.[9]

*(5-Chloro-2,4-dihydroxybenzoyl)-iso-indoline (**4**).* Yield 44%; white solid; mp 210−212 °C (petroleum spirits/Et_2O 2:1); R_f 0.38 (petroleum spirits/Et_2O 1:1); IR 3265, 1599, 1564 cm^{-1}; ^1H NMR (DMSO-d_6, 400 MHz) δ 10.42 (s, 1H), 10.33 (s, 1H), 7.36 (br m, 4H), 7.22 (s, 1H), 6.60 (s, 1H), 4.74 (app 2s, 2H + 2H); ^{13}C NMR (DMSO-d_6, 101 MHz) δ 166.7, 154.7, 154.1, 136.8, 135.9, 128.7 (2C), 127.2 (2C), 122.8, 116.3, 109.9, 103.8, 52.7, 51.7; HRMS (ESI-TOF) m/z: $[M + H]^+$ Calcd for $C_{15}H_{12}ClNO_3$ 298.1443; Found 298.1438; Anal. Calcd. for $C_{15}H_{12}ClNO_3$: C, 62.19; H, 4.17; N, 4.83. Found: C, 62.35; H, 4.10; N, 4.94.[7]

*N-(2,4-Dihydroxy-5-iso-propylbenzoyl)(5-((4-methylpiperazin-1-yl)methyl)-iso-indoline (**7**).* Resorcylate **8** (35 mg, 0.15 mmol) and *iso*-indoline trihydrochloride **26** (51 mg, 0.15 mmol) in THF (0.75 mL) were cooled to −20 °C, when ^{i}PrMgCl in THF (2.0 M; 0.45 mL, 0.90 mmol) was added dropwise with stirring. The mixture was warmed up to 0 °C over 3 h, and subsequently to room temperature over 12 h, when reaction was quenched with saturated aqueous NH_4Cl (10 mL). The aqueous layer was acidified to pH 6−7 using aqueous HCl (1.0 M) and extracted with EtOAc and CH_2Cl_2 (1:1; 30 mL). The organic layer was washed with brine (15 mL), dried ($MgSO_4$), the solvent evaporated under vacuum and the residue chromatographed (CH_2Cl_2/MeOH 19:1 to 4:1) to give the desired amide **7** (36 mg, 70%) as a white solid: mp 93−94 °C (acetone/MeOH 3:1); R_f 0.20 (CH_2Cl_2/MeOH 4:1); IR 3134, 1700, 1610, 1564 cm^{-1}; ^1H NMR ($CDCl_3$, 400 MHz) δ 7.59 (s, 1H), 7.34−7.31 (m, 2H), 7.28−7.26 (m, 1H), 6.42 (s, 1H), 5.08 (br s, 4H), 3.49 (s, 2H), 3.26 (sep, J = 2.8 Hz, 1H), 2.44 (br s, 8H), 2.23 (s, 3H), 1.26 (s, 6H); ^{13}C NMR ($CDCl_3$, 101 MHz) δ 172.6, 162.1, 160.4, 140.2, 138.5, 137.0, 130.2, 128.4, 127.3, 124.9, 124.2, 110.8, 104.7, 64.2, 56.7 (3C), 54.5 (3C), 47.0, 28.4, 24.0 (2C); HRMS (ESI-TOF) m/z: $[M + H]^+$ Calcd for $C_{24}H_{31}N_3O_3$ 410.2444; Found 410.2434.[9]

■ ASSOCIATED CONTENT

Ⓢ Supporting Information

Copies of ^1H and ^{13}C NMR spectra corresponding to all reported compounds. This material is available free of charge via the Internet at http://pubs.acs.org.

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Notes

The authors declare no competing financial interest.

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