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MEDICINAL CHEMISTRY RESEARCH

ORIGINAL RESEARCH

Synthesis, cytotoxicity and pro-apoptosis activity of isoquinoline quinones

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Abstract Mansouramycins are newly isolated cytotoxic isoquinoline quinones from marine organism. To find novel anticancer agents, eighteen isoquinoline quinones **7a–7r** as Mansouramycins analogs were designed and synthesized. Most of these compounds displayed moderate cytotoxicities against MCF-7, A549, HCT116, and HepG2 cancer cell lines at micromolar concentration. Compound **7a** was found to has the ability of inducing HepG2 cells apoptosis by Hoechst33342 staining assay.

Keywords Mansouramycins · Isoquinoline quinones · Synthesis · Cytotoxicity · Apoptosis

Introduction

Despite the rapid development of target-specific anticancer agents (Ling et al. 2014, 2015; Li et al. 2015), natural derived compounds still play important roles in cancer chemotherapy. Marine isoquinoline quinones alkaloids have received lots of attention from the scientific community due to their various biological activities (Hawas et al. 2009; Pettit et al. 2000; Milanowski et al. 2004; Li et al. 2017; Abdelwahab et al. 2014). These isoquinoline quinones, including the Caulibugulones, Cribrostatins, and Mansouramycins, were isolated from the marine sponges, marine bryozoans and marine streptomycetes (Choi et al. 1993; Brisson et al. 2007) (Fig. 1). Mansouramycin D showed cytotoxicity against 36 cancer cell lines with a mean IC_{50} value up to 0.089 μM (Hawas et al. 2009). Cribrostatin **5** showed proliferation inhibitory to six cancer cell lines with GI_{50} value from 45 to 360 nM (Brisson et al. 2007). These results suggested potential value of isoquinoline quinones as lead structures for the development of new anticancer agents (Vasquez et al. 2010). The total synthesis of Mansouramycin D have been completed involving intramolecular iminoannulation as a key ring closure step (Prakash and Nagarajan 2014). However, to the best of our knowledge, there is no structure–activity relationship (SAR) discussion on isoquinoline quinones. To fully understand the influence of C-3 substitute groups R_1 and C-7 substitute groups R_2 on their cytotoxicities, we intend to design and synthesize series of isoquinoline quinones (Scheme 1). In this paper, we report the synthsis of these isoquinoline quinones **7a–7r**, their cytotoxicities against four cancer cell lines. The effects of substitute groups R_1 and R_2 on their cytotoxicities. Most of these compounds displayed moderate cytotoxicities against MCF-7, A549, HCT116, and HepG2 cancer cell lines at micromolar concentration. Compound **7k** showed the best cytotoxicities among these compounds with IC_{50} values from 2.3 to 2.6 μM.

Results and discussion

Chemistry

Synthesis of isoquinoline quinones by various methods have been reported to date (Vasquez et al. 2009; Ferreira et al. 2003;

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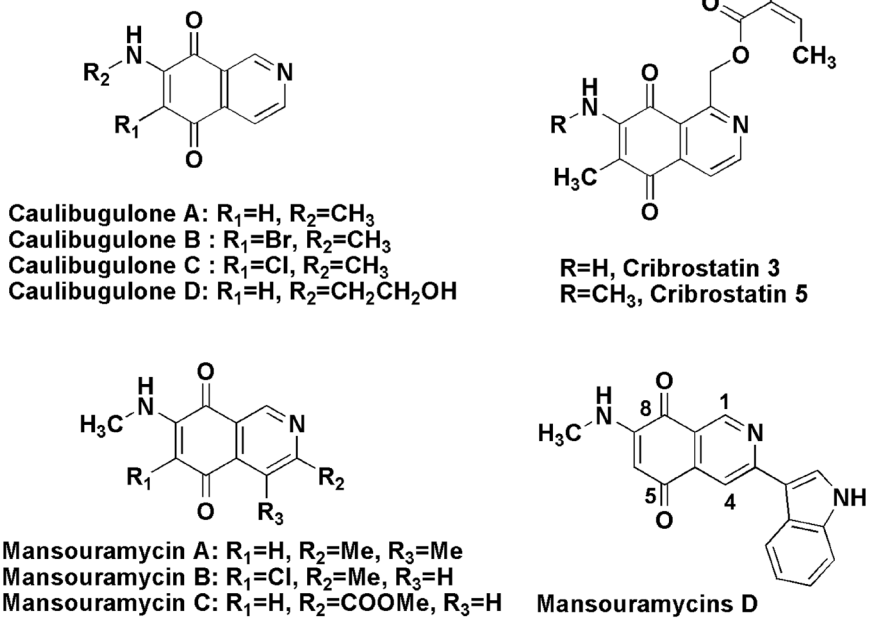
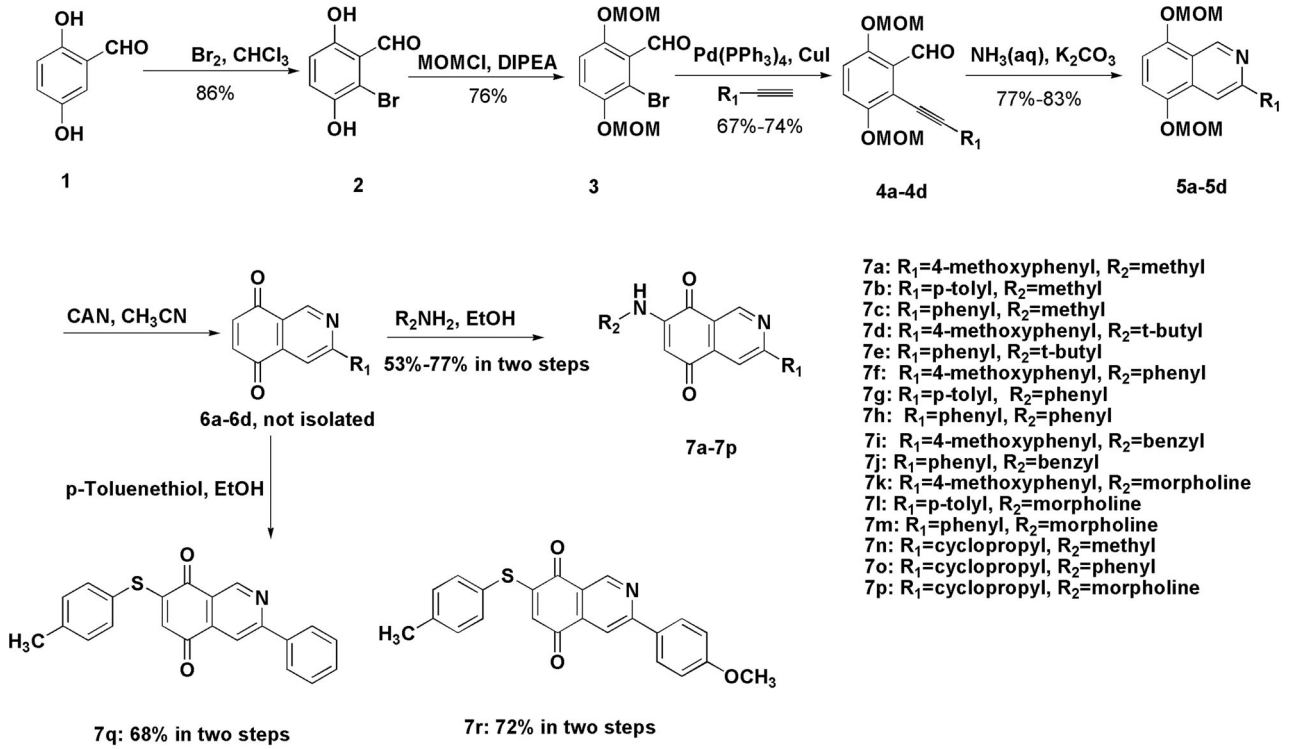
Fig. 1 Marine derived isoquinoline quinones





Scheme 1 Synthesis of isoquinoline quinines **7a–7r**

Perez et al. 2000; Coppola et al. 2015; Zhang et al. 2016; Prakash and Nagarajan 2015). Firstly, bromination of commercially available 2,5-dihydroxybenzaldehyde **1** afforded 2-bromo-3,5-dihydroxybenzaldehyde **2** in 86% yield. Then, both hydroxyl groups of 2-bromo-3,6-dihydroxybenzaldehyde were protected by a Methoxymethyl (MOM) group using MOM chloride and diisopropylethylamine as a base to afford compound **3** in 76% yield. The Sonogashira reaction with **3** and alkyne using $Pd(PPh_3)_4$ (5 mol%), CuI (5 mol%), and N,N-Dimethyl formamide (DMF) as solvent proceeded smoothly at 100 °C and yielded 67–74% of coupled products **4a–4d** (Scheme 1). The iminoannulation of **4a–4d** was performed

with aqueous ammonia and potassium carbonate to give **5a–5d** in 77–83% yields. Subsequently, oxidation and deprotection reactions of compounds **5a–5d** using ammonium ceric nitrate in one pot to afford **6a–6d**. Nextly, without further purification, compounds **6a–6d** were allowed to react with amines or thiophenols to give final eighteen products **7a–7r** in 53–77% yields.

Biological evaluation

Cytotoxicities

With eighteen isoquinoline quinones in hand, four cancer cell lines from various solid tumors, MCF-7, HCT-116, HepG2, and A549 were used to preliminaryly screen their cytotoxicities employing a 3-(4,5-Dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay by using etoposide as control (Table 1). IC_{50} values were summarized in Table 1 and represented the concentration inducing a 50% decrease of cell growth after 3 days incubation. From the IC_{50} values, it is clear that most of these compounds displayed cytotoxic effects on four cancer cells at micromolar concentration.

Table 1 In vitro cytotoxicities of compounds **7a–7r**

Compd	$IC_{50} \pm SD$ (μM)				
	MCF7	A549	HCT116	HepG2	Average[a]
7a	7.2 ± 0.4	3.6 ± 0.3	3.3 ± 0.1	1.9 ± 0.1	4.0 ± 0.3
7b	5.7 ± 0.6	5.5 ± 0.4	5.1 ± 0.3	2.5 ± 0.3	4.7 ± 0.1
7c	4.6 ± 0.1	2.7 ± 0.4	2.8 ± 0.1	4.0 ± 0.1	3.5 ± 0.4
7d	44.2 ± 2.1	14.4 ± 2.4	10.1 ± 1.6	15.2 ± 1.7	20.9 ± 1.4
7e	11.9 ± 1.0	25.5 ± 2.4	12.1 ± 1.2	6.3 ± 0.3	13.9 ± 0.7
7f	16.8 ± 0.9	8.9 ± 0.6	17.4 ± 0.8	5.5 ± 0.3	12.1 ± 0.5
7g	12.7 ± 0.6	10.7 ± 0.6	5.7 ± 0.3	4.3 ± 0.2	8.3 ± 0.9
7h	8.3 ± 0.7	4.9 ± 0.3	5.7 ± 0.4	2.4 ± 0.2	5.3 ± 0.1
7i	46.1 ± 5.0	45.8 ± 5.4	12.1 ± 1.4	5.4 ± 3.4	27.3 ± 2.5
7j	0.7 ± 0.1	8.5 ± 0.7	6.9 ± 0.8	1.1 ± 0.1	4.3 ± 0.3
7k	2.6 ± 0.2	2.2 ± 0.1	2.2 ± 0.3	2.3 ± 0.2	2.3 ± 0.1
7l	6.6 ± 0.4	6.6 ± 0.3	6.8 ± 0.4	4.8 ± 0.5	6.2 ± 0.4
7m	7.1 ± 0.6	6.7 ± 0.8	7.1 ± 0.5	8.2 ± 0.6	7.3 ± 0.3
7n	8.5 ± 0.5	8.7 ± 0.4	5.8 ± 0.6	6.2 ± 0.6	7.3 ± 0.9
7o	20.4 ± 2.4	20.7 ± 1.7	5.9 ± 0.3	6.9 ± 0.4	13.4 ± 0.9
7p	7.1 ± 0.5	9.2 ± 0.7	6.1 ± 0.8	4.0 ± 0.7	6.6 ± 0.5
7q	34.6 ± 3.1	37.1 ± 2.5	26.6 ± 2.0	10.0 ± 1.1	27.1 ± 2.4
7r	>100	>100	>100	27.6	>100
Etoposide	4.8 ± 0.3	11.3 ± 0.1	8.7 ± 0.5	4.9 ± 0.1	7.4 ± 0.3

All experiments were independently performed at least three times

[a] Average IC_{50} of four cancer cells IC_{50}s

The C-3 position of isoquinoline quinines scaffold proved to be critical for their cytotoxicities. The introducing of cyclopropyl group at C-3 position of **7n** and **7o** resulted in 2–3-folds decrease in their cytotoxicities compared to compound **7a**, which suggested a phenyl group at C-3 position was favorable to their cytotoxicities. For compounds **7a**, **7b**, and **7c** having a methylamino groups at C-7 position, they showed almost the same cytotoxicities with the average IC_{50} values 4.0, 4.7, and 3.5 μM, which indicated that substituted patterns on C-3 phenyl group have no influence on their cytotoxicities. For compounds **7f**, **7g**, **7h**, and **7o** bearing a benzylamino groups at C-7 position, the substituted groups at C-3 position have influence on their cytotoxicities. Compound **7h** with benzyl group at a C-3 position exhibited the best cytotoxicity with the average IC_{50} values 5.3 μM, while compound **7o** with cyclopropyl group at a C-3 position possessed the worst cytotoxicity with the average IC_{50} values 13.4 μM. For compounds **7c**, **7e**, **7h, 7j**, and **7m** bearing phenyl group at C-3 position, **7c** showed the best cytotoxicities with IC_{50} values 3.5 μM, which indicated small substituted group was favorable to their cytotoxicities.

In general, compounds **7a** and **7k** displayed the best cytotoxicities among these compounds with IC_{50} values from 1.9 to 7.2 μM, which were better than those of etoposide. Compounds **7q** and **7r** which bearing sulfur ether groups showed less cytotoxicities than amines, which indicated amino-substitution was favorable to their cytotoxicities. The amino-substitutions had practically significant effects on their cytotoxicities, methylamine, and morpholine substitute groups were favorable to the cytotoxicities. These compounds showed no selectivity for these four cancer cell lines.

Apoptosis inducing assay

On the basis of the above results, further biological evaluations have been focused on compound **7a** HepG2 cells were stained with Hoechst33342 to verify the type of cell death. Without compound **7a** treatment, the nuclei of control cells showed uniform blue fluorescence, which indicated the cells were healthy and the nuclei were intact (Fig. 2a). However, after treatment with different concentration of compound **7a** (0.1, 5, and 10 μM) for 48 h, the number of cells was significantly reduced, and most cells showed early apoptosis with perinuclear chromatin condensation and late apoptosis with nuclear dense chromatin of pneumocytes (Fig. 2a). These results showed that **7a** induced the cells death occurred primarily through apoptosis in a dose-dependent manner. The data collected from the manual counting of cells with normal and apoptotic nuclear features are shown in Fig. 2b.

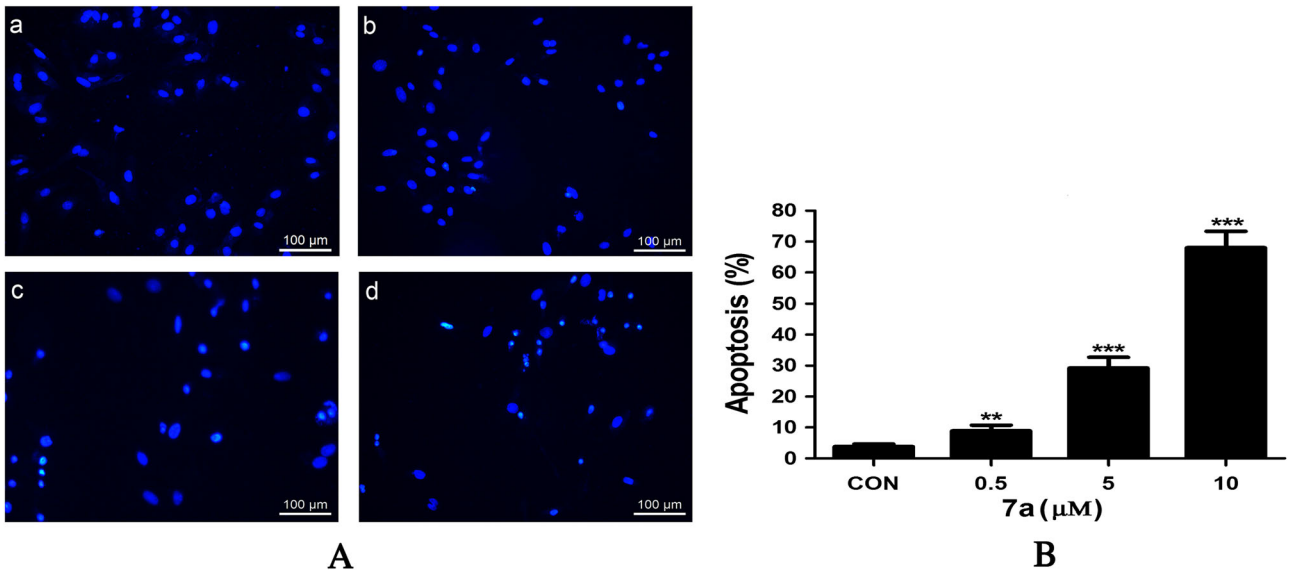


Fig. 2 Nuclear morphology of HepG2 Cells Treated with Compound 7a in 0, 0.5, 5, and 10 μM, respectively for 48 h by Hoechst33342 staining

Experimental

Chemistry

Melting points (mp) were determined with a X5 melting point apparatus (Yuhua, Gongyi, China). Commercial grade reagents and solvents were used without further purification, except where noted. Flash chromatography was performed on silica gel (100–200 mesh, Yinlong, Yantai, China). Thin layer chromatography (TLC) was performed on precoated Merck silica Gel 60 F_{254} plates (Merck, Darmstadt, Germany). ^{1}H nuclear magnetic resonance (NMR) spectra were recorded in $CDCl_3$ solution (if not otherwise stated) with a Bruker DRX-400 instrument at 400 MHz, with Me_4Si as internal standard. ^{13}C NMR spectra were recorded with the same instrument at 100 MHz under the same conditions. Full scan mass spectra of the compounds were acquired in the range 50–800 m/z with an Agilent 500MS Ion trap mass spectrometer equipped with an electrospray ionization source.

Compound **3** was prepared according to the literature procedure from commercially available 2,5-dihydroxybenzaldehyde **1** in 65% yields in two steps (Little and Porco 2012).

Synthetic procedure for synthesis of 4a–4d

*3,6-Bis-methoxymethoxy-2-(4-methoxy-phenylethynyl)-benzaldehyde (**4a**)*

To a solution of compound **3** (1.0 g, 3.27 mmol) in DMF (5 mL) was added CuI (32 mg, 0.16 mmol), $Pd(PPh_3)_4$ (189 mg, 0.16 mmol) and Na_2CO_3 (5.19 g, 4.91 mmol) under nitrogen atmosphere. Then 4-methoxyphenylacetylene (648 mg, 4.91 mmol) was added slowly, and the mixture was heated at 100 °C for 8 h. After completion of reaction (indicated by TLC), reaction mixture was diluted with 50 mL of $CHCl_3$ and filtered with celite bed. Then, water (10 mL) was added to the diluted solution, which was then extracted with $CHCl_3$ (2 × 50 mL). The combined organic layer was dried with anhydrous Na_2SO_4 and concentrated under vacuum and purified by column chromatography on silica gel eluting with EtOAc/petroleum-ether (1:6 v/v) to afford **4a** in 71% yield.

Dark brown solid; mp 64–66 °C; ^{1}H NMR ($CDCl_3$, 400 MHz) $\delta = 10.67$ (s, 1H, CHO), 7.54 (d, $J = 8.0$ Hz, 2H, ArH), 7.28 (d, $J = 8.0$ Hz, 1H, ArH), 7.13 (d, $J = 8.0$ Hz, 1H, ArH), 6.89 (d, $J = 8.0$ Hz, 2H, ArH), 5.23–5.21 (m, 4H, $-CH_2-$), 3.83 (s, 3H, $-OCH_3$), 3.55(s, 3H, $-CH_3$), 3.51(s, 3H, $-CH_3$); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 190.3$ (C=O), 154.1(ArC), 153.3(ArC), 139.2(ArC), 132.2(ArC), 132.1(ArC), 129.3(ArC), 129.2(ArC), 126.4(ArC), 122.5 (ArC), 119.6(ArC), 117.3(ArC), 116.5(ArC), 100.2 (OCH_2O), 96.1(OCH_2O), 95.5(C≡C), 81.9(C≡C), 56.5 (OCH_3), 56.4(OCH_3), 55.6(OCH_3); low resolution mass spectrometry (LRMS) (ESI-TOF) (*m/z*) $[M+H]^+$ calcd for $C_{20}H_{20}O_6$ 356.1, found 356.1.

*3,6-Bis-methoxymethoxy-2-p-tolylethynyl-benzaldehyde (**4b**)*

Dark brown solid, yield 72%; mp 71–74 °C; ^{1}H NMR ($CDCl_3$, 400 MHz) $\delta = 10.67$ (s, 1H, CHO), 7.49 (d, $J = 8.0$ Hz, 2H, ArH), 7.27 (d, $J = 8.0$ Hz, 2H, ArH), 7.17(d, $J = 8.0$ Hz, 2H, ArH), 5.24 (s, 4H, $-CH_2-$), 3.55(s, 3H, $-CH_3$), 3.51(s, 3H, $-CH_3$), 2.38 (s, 3H, $-CH_3$); ^{13}C NMR

($CDCl_3$, 100 MHz): $\delta = 190.3$(C=O), 153.3(ArC), 153.2 (ArC), 139.1(ArC), 131.7(ArC), 131.7(ArC), 129.2(ArC), 129.2(ArC), 126.6(ArC), 122.5(ArC), 119.9(ArC), 117.1 (ArC), 116.7(ArC), 100.6(OCH_2O), 96.0(OCH_2O), 95.5 (C≡C), 81.9(C≡C), 56.5(OCH_3), 56.4(OCH_3), 21.6(CH_3); LRMS (ESI-TOF) (*m/z*) $[M+H]^+$ calcd for $C_{20}H_{20}O_5$ 340.1, found 340.1.

*3,6-Bis-methoxymethoxy-2-phenylethynyl-benzaldehyde (**4c**)*

Dark brown oil, yield 74%; ^1H NMR ($CDCl_3$, 400 MHz) $\delta = 10.67$ (s, 1H, CHO), 7.60 (d, $J = 8.0$ Hz, 2H, ArH), 7.37–7.35 (m, 3H, ArH), 7.30(d, $J = 8.0$ Hz, 1H, ArH), 7.16 (d, $J = 8.0$ Hz, 1H, ArH), 5.24 (s, 4H, –CH_2–), 3.56(s, 3H, –CH_3), 3.51(s, 3H, –CH_3); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 190.3$(C=O), 153.3(ArC), 153.0(ArC), 139.2(ArC), 131.7(ArC), 131.6(ArC), 129.2(ArC), 129.1(ArC), 126.3 (ArC), 122.1(ArC), 119.9(ArC), 117.13(ArC), 117.1(ArC), 100.6(OCH_2O), 96.0(OCH_2O), 95.4(C≡C), 82.1(C≡C), 56.5(OCH_3), 56.4(OCH_3); LRMS (ESI-TOF) (*m/z*) $[M+H]^+$ calcd for $C_{19}H_{18}O_5$ 326.1, found 326.1.

*2-Cyclopropylethynyl-3,6-bis-methoxymethoxy-benzaldehyde (**4d**)*

Dark brown oil, yield 67%; ^1H NMR ($CDCl_3$, 400 MHz): $\delta = 10.57$ (s, 1H, CHO), 7.31–7.33 (d, $J = 8.0$ Hz, 1H, ArH), 7.12–7.14 (d, $J = 8.0$ Hz, 1H, ArH), 5.21 (s, 4H, –CH_2–), 3.52(s, 3H, –CH_3), 3.50(s, 3H, –CH_3), 3.50(m, 1H, –CH–), 0.92–0.94(m, 4H, –CH_2–); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 192.2$(C=O), 166.0(ArC), 150.4(ArC), 126.7 (ArC), 122.4(ArC), 121.6(ArC), 116.0(ArC), 114.6 (OCH_2O), 95.9(OCH_2O), 95.4(C≡C), 95.2(C≡C), 56.4 (OCH_3), 56.4(OCH_3), 10.6(CH_2), 9.2(CH_2), 0.89(CH); LRMS (ESI-TOF) (*m/z*) $[M+H]^+$ calcd for $C_{16}H_{18}O_5$ 290.1, found 290.1.

Synthetic procedure for synthesis of 5a–5d

*5,8-Bis-methoxymethoxy-3-(4-methoxy-phenyl)-isoquinoline (**5a**)*

An oven-dried 25 mL round-bottomed flask equipped with a Teflon-coated magnetic stirring bar was charged with **4a** (600 mg, 1.68 mmol) and K_2CO_3 (468 mg, 3.36 mmol), 10 mL of ethanol and 3 mL of aqueous ammonia (37% ammonia in water). The reaction mixture was refluxed for 8 h. The complete conversion of starting material was observed by TLC. The reaction was allowed to cool to room temperature, poured into ice and extracted with $CHCl_3$ (10 mL). The organic layer was washed with 10 mL of water and 5 mL of brine, dried over anhydrous sodium sulfate and solvent was removed under reduced pressure. The crude product was purified by column chromatography (EtOAc/petroleum-ether 1:10 v/v) to afford **5a** in 83% yield.

Dark brown solid; mp 107–109 °C; ^1H NMR ($CDCl_3$, 400 MHz): $\delta = 9.64$ (s, 1H, ArH), 8.30 (s, 1H, ArH), 8.12 (d, $J = 8.0$ Hz, 2H, ArH), 7.17(d, $J = 8.0$ Hz, 1H, ArH), 7.05–7.03(m, 3H, ArH), 5.36 (s, 4H, –CH_2–), 3.88(s, 3H, –OCH_3), 3.57–3.54(m, 6H, –OCH_3); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 158.8$(ArC), 151.6(ArC), 148.6(ArC), 147.3 (ArC), 146.7(ArC), 138.6(ArC), 136.9(ArC), 129.6(ArC), 129.5(ArC), 126.9(ArC), 126.8(ArC), 120.4(ArC), 112.6 (ArC), 110.2(ArC), 108.7(ArC), 95.2(OCH_2O), 95.1 (OCH_2O), 56.4(OCH_3), 56.3(OCH_3), 55.3(OCH_3); LRMS (ESI-TOF) (*m/z*) $[M+H]^+$ calcd for $C_{20}H_{21}NO_5$ 355.1, found 355.1.

*5,8-Bis-methoxymethoxy-3-p-tolyl-isoquinoline (**5b**)*

Brown solid; yield 80%; mp 112–113 °C; ^1H NMR ($CDCl_3$, 400 MHz): $\delta = 9.69$ (s, 1H, ArH), 8.39 (s, 1H, ArH), 8.10 (d, $J = 8.0$ Hz, 2H, ArH), 7.35(d, $J = 8.0$ Hz, 2H, ArH), 7.21(d, $J = 8.0$ Hz, 1H, ArH), 7.06(d, $J = 8.0$ Hz, 1H, ArH), 5.40–5.38 (m, 4H, –CH_2–), 3.59–3.58(m, 6H, –CH_3), 2.46 (s, 3H, –CH_3); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 151.6$ (ArC), 148.5(ArC), 147.3(ArC), 146.7(ArC), 138.5(ArC), 136.9(ArC), 130.5(ArC), 129.6(ArC), 129.5(ArC), 126.9 (ArC), 126.8(ArC), 120.4(ArC), 112.6(ArC), 110.2(ArC), 108.7(ArC), 95.2(OCH_2O), 95.1(OCH_2O), 56.4(OCH_3), 56.3(OCH_3), 21.3(CH_3); LRMS (ESI-TOF) (*m/z*) $[M+H]^+$ calcd for $C_{20}H_{21}NO_4$ 339.1, found 339.1.

*5,8-Bis-methoxymethoxy-3-phenyl-isoquinoline (**5c**)*

Brown solid; yield 83%; mp 78–80 °C; ^1H NMR ($CDCl_3$, 400 MHz): $\delta = 9.68$ (s, 1H, ArH), 8.39 (s, 1H, ArH), 7.52 (d, $J = 8.0$ Hz, 2H, ArH), 7.20(d, $J = 8.0$ Hz, 2H, ArH), 7.07–7.05(m, 3H, ArH), 5.38–5.36 (m, 4H, –CH_2–), 3.55–3.57(m, 6H, –CH_3); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 151.6$(ArC), 148.6(ArC), 147.4(ArC), 146.5(ArC), 138.6(ArC), 136.9(ArC), 129.6(ArC), 129.5(ArC), 127.2 (ArC), 126.9(ArC), 126.8(ArC), 120.4(ArC), 112.6(ArC), 110.2(ArC), 108.7(ArC), 95.2(OCH_2O), 95.1(OCH_2O), 56.4(OCH_3), 56.3(OCH_3); LRMS (ESI-TOF) (*m/z*) $[M+H]^+$ calcd for $C_{19}H_{19}NO_4$ 325.1, found 325.1.

*5,8-Bis-methoxymethoxy-3-cyclopropyl-isoquinoline (**5d**)*

Brown solid; yield 77%; mp 115–117 °C;^1H NMR ($CDCl_3$, 400 MHz): $\delta = 9.48$ (s, 1H, ArH), 7.78 (s, 1H, ArH), 6.94 (d, $J = 8.0$ Hz, 2H, ArH), 5.35–5.33 (m, 4H, –CH_2–), 3.55–3.53(m, 6H, –CH_3), 2.23–2.21(m, 1H, –CH–), 1.13–1.12(m, 2H, –CH_2–), 1.06–1.04(m, 2H, –CH_2–); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 156.6$(ArC), 148.5(ArC), 147.0(ArC), 145.9(ArC), 130.2(ArC), 119.9(ArC), 112.4

(ArC), 110.5(ArC), 107.5(ArC), 95.2(OCH_2O), 94.9 (OCH_2O), 56.3(OCH_3), 56.2(OCH_3), 17.3(CH), 9.5(CH_2), 9.4(CH_2); LRMS (ESI-TOF) (*m/z*) $[M + H]^+$ calcd for $C_{16}H_{19}NO_4$ 289.1, found 289.1.

Synthetic procedure for synthesis of 6a–6d

To a solution of **5a** (150 mg, 0.42 mmol) in CH_3CN (5 mL) was added CAN (690 mg, 1.26 mmol). The mixture was allowed to stir 2 h at room temperature. Then, EtOAc (10 mL) was added. The mixture was washed with 10 mL of water and 5 mL of brine, dried over anhydrous sodium sulfate and solvent was removed under reduced pressure to give a brown oil **6a**. This crude material was then used for next step.

Synthetic procedure for synthesis of 7a–7r

*3-(4-Methoxy-phenyl)-7-methylamino-isoquinoline-5,8-dione (**7a**)*

The residue was diluted with 5 mL of 1,2-dimethoxymethane. The reaction mixture was then cooled to 0 °C and 33% wt. absolute ethanolic solution of methyl amine (1.35 mL, 1.26 mmol) was added dropwise. Then it was allowed to stir at room temperature. After complete conversion was observed in TLC. After removing the solvent in reduced pressure, the reaction mixture was poured in 10 mL of water and extracted with ethyl acetate (3 × 50 mL). The organic layer was washed with water (20 mL) and brine (20 mL), dried over anhydrous sodium sulfate and the solvent was removed under reduced pressure. The crude product was purified by column chromatography to give **7a** in 74% yield for two steps.

Red solid; mp 191–192 °C; ^{1}H NMR ($CDCl_3$, 400 MHz): $\delta = 9.24$ (s, 1H, ArH), 8.30 (s, 1H, ArH), 8.16 (d, $J = 8.0$ Hz, 2H, ArH), 7.03 (d, $J = 8.0$ Hz, 2H, ArH), 5.81 (s, 1H, ArH), 3.89 (s, 3H, –OCH_3), 2.97(s, 3H, –CH_3); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 181.3$(C=O), 180.7(C=O), 163.2 (N–C), 162.0(ArC), 149.3(ArC), 148.3(ArC), 140.0(ArC), 130.5(ArC), 129.3(ArC), 129.3(ArC), 128.9(ArC), 122.0 (ArC), 114.4(ArC), 114.3(ArC), 101.0(=CH), 55.4(OCH_3), 29.2(NCH_3); high resolution mass spectrum (HRMS) (ESI-TOF) (*m/z*) $[M + H]^+$ calcd for $C_{17}H_{14}N_2O_3$ 294.1004, found 294.1000.

*3-p-Tolyl-7-methylamino-isoquinoline-5,8-dione (**7b**)*

Red solid; yield 71%; mp 197–199 °C; ^{1}H NMR ($CDCl_3$, 400 MHz): $\delta = 9.30$ (s, 1H, ArH), 8.37 (s, 1H, ArH), 8.11 (d, $J = 8.0$ Hz, 2H, ArH), 7.35–7.33(m, 2H, ArH), 5.85(s, 1H, ArH), 2.99–2.97(m, 3H, –CH_3), 2.46(s, 3H, –CH_3); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 181.2$(C=O), 180.8(C=O), 163.6(N–C), 149.2(ArC), 148.3(ArC), 141.3(ArC), 140.1 (ArC), 135.1(ArC), 129.8(ArC), 129.7(ArC), 127.7(ArC), 127.6(ArC), 127.2(ArC), 115.2(ArC), 101.0(=CH), 29.3 (NCH_3), 21.5(CH_3); HRMS (ESI-TOF) (*m/z*) $[M + H]^+$ calcd for $C_{17}H_{14}N_2O_2$ 279.1128, found 279.1128

*3-Phenyl-7-methylamino-isoquinoline-5,8-dione (**7c**)*

Red solid; yield 77%; mp 184–186 °C; ^{1}H NMR ($CDCl_3$, 400 MHz): $\delta = 9.34$ (s, 1H, ArH), 8.41 (s, 1H, ArH), 8.21 (d, $J = 8.0$ Hz, 2H, ArH), 7.55–7.53 (m, 2H, ArH), 5.86(s, 1H, ArH), 3.00–3.01(d, $J = 8.0$ Hz, 3H, –CH_3); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 181.1$(C=O), 180.7(C=O), 163.6 (N–C), 149.2(ArC), 148.8(ArC), 148.3(ArC), 140.2(ArC), 137.8(ArC), 130.8(ArC), 129.1(ArC), 129.0(ArC), 127.7 (ArC), 127.6(ArC), 115.6(ArC), 101.1(=CH), 29.3(NCH_3); HRMS (ESI-TOF) (*m/z*) $[M + H]^+$ calcd for $C_{16}H_{12}N_2O_2$ 264.0899, found 264.0902

*3-(4-Methoxy-phenyl)-7-tert-butylamino-isoquinoline-5,8-dione (**7d**)*

Red solid; yield 67%; mp 207–209 °C; ^{1}H NMR ($CDCl_3$, 400 MHz): $\delta = 9.23$ (s, 1H, ArH), 8.28 (s, 1H, ArH), 8.16 (d, $J = 8.0$ Hz, 2H, ArH), 7.03(d, $J = 8.0$ Hz, 2H, ArH), 6.06(s, 1H, ArH), 3.89(s, 3H, –OCH_3), 1.47(s, 9H, –CH_3); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 181.3$(C=O), 181.0 (C=O), 163.0(N–C), 162.0(ArC), 148.4(ArC), 146.3(ArC), 139.8(ArC), 130.4(ArC), 129.3(ArC), 129.2(ArC), 129.1 (ArC), 122.0(ArC), 111.4(ArC), 111.3(ArC), 103.0(=CH), 55.5(OCH_3), 52.1($\underline{C}(CH_3)_3$), 28.2(3 × CH_3); HRMS (ESI-TOF) (*m/z*) $[M + H]^+$ calcd for $C_{20}H_{20}N_2O_3$ 336.1474, found 336.1477

*3-Phenyl-7-tert-butylaminoisoquinoline-5,8-dione (**7e**)*

Red solid; yield 69%; mp 203–204 °C; ^{1}H NMR ($CDCl_3$, 400 MHz): $\delta = 9.30$ (s, 1H, ArH), 8.37 (s, 1H, ArH), 8.19 (d, $J = 8.0$ Hz, 2H, ArH), 7.52 (d, $J = 8.0$ Hz, 2H, ArH), 6.08(s, 1H, ArH), 1.48(s, 9H, –CH_3); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 181.4$(C=O), 181.0(C=O), 163.5(N–C), 163.0(ArC), 148.3(ArC), 146.2(ArC), 140.0(ArC), 137.8 (ArC), 130.7(ArC), 129.1(ArC), 129.0(ArC), 127.6(ArC), 127.5(ArC), 122.7(ArC), 115.4(ArC), 103.1(=CH), 52.1($\underline{C}(CH_3)_3$), 28.3(3 × CH_3); HRMS (ESI-TOF) (*m/z*) $[M + H]^+$ calcd for $C_{19}H_{18}N_2O_2$ 306.1368, found 307.1436.

*3-(4-Methoxy-phenyl)-7-phenylamino-isoquinoline-5,8-dione (**7f**)*

Red solid; yield 53%; mp 199–201 °C; ^{1}H NMR ($CDCl_3$, 400 MHz): $\delta = 9.34$ (s, 1H, ArH), 8.30 (s, 1H, ArH), 8.18 (d, $J = 8.0$ Hz, 2H, ArH), 7.44 (d, $J = 8.0$ Hz, 1H, ArH),

7.31 (d, $J = 8.0$ Hz, 2H, ArH), 7.28–7.26 (m, 3H, ArH), 7.05 (d, $J = 8.0$ Hz, 2H, ArH), 6.48(s, 1H, ArH), 3.90(s, 3H, –OCH_3); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 182.7$ (C=O), 181.1(C=O), 162.1(N–C), 148.5(ArC), 145.3 (ArC), 141.4(ArC), 138.4(ArC), 136.1(ArC), 135.0(ArC), 129.2(ArC), 129.1(ArC), 129.0(ArC), 127.8(ArC), 127.6 (ArC), 127.5(ArC), 124.3(ArC), 121.5(ArC), 121.4(ArC), 114.4(ArC), 113.2(ArC), 103.5(=CH), 55.4(OCH_3); HRMS (ESI-TOF) (*m/z*) $[M + H]^+$ calcd for $C_{22}H_{16}N_2O_3$ 356.1161, found 356.1163.

*3-p-Tolyl-7-phenylamino-isoquinoline-5,8-dione (**7g**)*

Red solid; yield 62%; mp 207–209 °C; ^{1}H NMR ($CDCl_3$, 400 MHz): $\delta = 9.38$ (s, 1H, ArH), 8.36 (s, 1H, ArH), 8.13 (d, $J = 8.0$ Hz, 2H, ArH), 7.47 (d, $J = 8.0$ Hz, 2H, ArH), 7.37–7.35 (m, 3H, ArH), 7.34-7.32 (m, 2H, ArH), 6.51(s, 1H, ArH), 2.47(s, 3H, -CH_3); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 182.5$(C=O), 181.0(C=O), 163.8(N-C), 148.5(ArC), 145.3(ArC), 141.4(ArC), 139.5(ArC), 136.9(ArC), 135.0 (ArC), 130.2(ArC), 129.8(ArC), 129.5(ArC), 129.1(ArC), 127.6(ArC), 127.2(ArC), 126.3(ArC), 123.3(ArC), 123.0 (ArC), 114.9(ArC), 114.6(ArC), 103.5(=CH), 22.5(CH_3); HRMS (ESI-TOF) (*m/z*) $[M+H]^+$ calcd for $C_{22}H_{16}N_2O_2$ 340.1212, found 341.1281.

*3-Phenyl-7-phenylamino-isoquinoline-5,8-dione (**7h**)*

Red solid; yield 65%; mp 183–185 °C; ^{1}H NMR ($CDCl_3$, 400 MHz): $\delta = 9.39$ (s, 1H, ArH), 8.38 (s, 1H, ArH), 8.20 (d, $J = 8.0$ Hz, 2H, ArH), 7.53 (d, $J = 8.0$ Hz, 3H, ArH), 7.48–7.46 (m, 2H, ArH), 7.31–7.29 (m, 3H, ArH), 6.49(s, 1H, ArH); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 182.3$(C=O), 181.1(C=O), 163.8(N–C), 148.5(ArC), 145.3(ArC), 139.6 (ArC), 137.8(ArC), 136.9(ArC), 130.9(ArC), 129.8(ArC), 129.5(ArC), 129.3(ArC), 129.0(ArC), 127.7(ArC), 127.3 (ArC), 126.3(ArC), 123.5(ArC), 123.0(ArC), 122.5(ArC), 115.2(ArC), 103.6(=CH); HRMS (ESI-TOF) (*m/z*) $[M + H]^+$ calcd for $C_{21}H_{14}N_2O_2$ 326.1055, found 326.1059

*3-(4-Methoxy-phenyl)-7-benzylamino-isoquinoline-5,8-dione (**7i**)*

Red solid; yield 71%; mp 205–207 °C; ^{1}H NMR ($CDCl_3$, 400 MHz): $\delta = 9.26$ (s, 1H, ArH), 8.28 (s, 1H, ArH), 8.16 (d, $J = 8.0$ Hz, 2H, ArH), 7.40–7.38 (m, 2H, ArH), 7.36–7.34 (m, 2H, ArH), 7.26 (s, 1H, ArH), 7.03 (d, $J = 8.0$ Hz, 2H, ArH), 5.87 (s, 1H, ArH), 4.41 (d, $J = 8.0$ Hz, 2H, –CH_2–), 3.89(s, 3H, –OCH_3); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 181.5$(C=O), 180.7(C=O), 163.2(N–C), 162.0 (ArC), 148.4(ArC), 148.1(ArC), 139.6(ArC), 135.5(ArC), 130.4(ArC), 129.7(ArC), 129.5(ArC), 129.3(ArC), 129.1 (ArC), 128.9(ArC), 128.3(ArC), 127.7(ArC), 127.3(ArC), 122.0(ArC), 114.4(ArC), 114.1(ArC), 102.0(=CH), 55.5 (OCH_3), 46.9($PhCH_2$); HRMS (ESI-TOF) (*m/z*) $[M + H]^+$ calcd for $C_{23}H_{18}N_2O_3$ 370.1317, found 370.1322.

*3-Pheny-7-benzylamino-l-isoquinoline-5,8-dione (**7j**)*

Red solid; yield 75%; mp 197–198 °C; ^{1}H NMR ($CDCl_3$, 400 MHz): $\delta = 9.31$ (s, 1H, ArH), 8.36 (s, 1H, ArH), 8.18 (d, $J = 8.0$ Hz, 2H, ArH), 7.53–7.51 (m, 3H, ArH), 7.38–7.37 (m, 2H, ArH), 7.36–7.34 (m, 2H, ArH), 7.26 (s, 1H, ArH), 5.89(s, 1H, ArH), 4.41 (d, $J = 4.0$ Hz, 2H, –CH_2–); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 181.4$(C=O), 180.1(C=O), 163.7(N–C), 148.4(ArC), 148.0(ArC), 140.0 (ArC), 137.8(ArC), 135.4(ArC), 130.8(ArC), 122.7(ArC), 129.7(ArC), 129.2(ArC), 129.0(ArC), 128.7(ArC), 128.4 (ArC), 127.7(ArC), 127.3(ArC), 127.0(ArC), 126.6(ArC), 115.4(ArC), 102.1(=CH), 46.9($PhCH_2$); HRMS (ESI-TOF) (*m/z*) $[M + H]^+$ calcd for $C_{22}H_{17}N_2O_2$ 341.1290, found 340.1209.

*3-(4-Methoxy-phenyl)-7-morpholin-4-yl-isoquinoline-5,8-dione (**7k**)*

Red solid; yield 64%; mp 219–221 °C; ^{1}H NMR ($CDCl_3$, 400 MHz): $\delta = 9.24$ (s, 1H, ArH), 8.23 (s, 1H, ArH), 8.16 (d, $J = 8.0$ Hz, 2H, ArH), 7.04 (d, $J = 8.0$ Hz, 2H, ArH), 6.10(s, 1H, ArH), 3.94–3.92 (m, 2H, –CH_2–), 3.90–3.89 (m, 2H, –CH_2–), 3.89 (s, 3H, –OCH_3), 3.63–3.61 (m, 4H, $2 \times$ –CH_2–); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 182.4$ (C=O), 182.0(C=O), 162.3(N–C), 161.9(ArC), 153.4 (ArC), 148.9(ArC), 138.5(ArC), 130.3(ArC), 129.2(ArC), 129.0(ArC), 123.9(ArC), 114.6(ArC), 114.2(ArC), 113.4 (ArC), 111.2(=CH), 66.5(CH_2), 66.3(CH_2), 55.5(OCH_3), 49.2(CH_2), 49.0(CH_2); HRMS (ESI-TOF) (*m/z*) $[M+H]^+$ calcd for $C_{20}H_{19}N_2O_4$ 351.1345, found 351.1334.

*3-p-Tolyl-7-morpholin-4-yl-isoquinoline-5,8-dione (**7l**)*

Red solid; yield 69%; mp 213–214 °C; ^{1}H NMR ($CDCl_3$, 400 MHz): $\delta = 9.26$ (s, 1H, ArH), 8.26 (s, 1H, ArH), 8.07 (d, $J = 8.0$ Hz, 2H, ArH), 7.33 (d, $J = 8.0$ Hz, 2H, ArH), 6.10(s, 1H, ArH), 3.89–3.87 (m, 4H, $2 \times$ –CH_2–), 3.62–3.60 (m, 4H, $2 \times$ –CH_2–), 2.45 (s, 3H, –CH_3); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 182.4$(C=O), 182.0(C=O), 162.7(N–C), 153.3(ArC), 148.8(ArC), 141.2(ArC), 138.5 (ArC), 134.9(ArC), 129.8(ArC), 129.6(ArC), 127.5(ArC), 127.3(ArC), 124.4(ArC), 114.1(ArC), 111.2(=CH), 66.4 (CH_2), 66.1(CH_2), 49.2(CH_2), 49.0(CH_2), 21.3(CH_3); HRMS (ESI-TOF) (*m/z*) $[M + H]^+$ calcd for $C_{20}H_{19}N_2O_3$ 335.1396, found 335.1386.

*3-Phenyl-7-morpholin-4-yl-isoquinoline-5,8-dione (**7m**)*

Red solid; yield 71%; mp 188–189 °C; ^{1}H NMR ($CDCl_3$, 400 MHz): $\delta = 9.29$ (s, 1H, ArH), 8.31 (s, 1H, ArH), 8.18 (d, $J = 8.0$ Hz, 2H, ArH), 7.55–7.53 (m, 3H, ArH), 6.12(s, 1H, ArH), 3.90–3.89(m, 4H, 2 × –CH_2–), 3.64–3.63 (m, 4H, 2 × –CH_2–); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 182.3$ (C=O), 182.1(C=O), 162.8(N–C), 153.3(ArC), 148.8 (ArC), 138.6(ArC), 137.8(ArC), 130.7(ArC), 129.3(ArC), 129.1(ArC), 127.6(ArC), 127.3(ArC), 124.6(ArC), 114.5 (ArC), 111.3(=CH), 66.4(CH_2), 66.1(CH_2), 49.2(CH_2), 49.0(CH_2); HRMS (ESI-TOF) (*m/z*) $[M+H]^+$ calcd for $C_{19}H_{17}N_2O_3$ 321.1239, found 321.1233.

*3-Cyclopropyl-7-methylamino-isoquinoline-5,8-dione (**7n**)*

Red solid; yield 67%; mp 123–125 °C; ^{1}H NMR ($CDCl_3$, 400 MHz): $\delta = 9.08$ (s, 1H, ArH), 7.73 (s, 1H, ArH), 5.79 (s, 1H, ArH), 2.96 (d, $J = 8.0$ Hz, 3H, –CH_3), 2.26 (m, 1H, –CH–), 1.26-1.22 (m, 4H, 2 × –CH_2–); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 181.5$(C=O), 180.8(C=O), 171.9(N–C), 149.2(ArC), 148.0(ArC), 139.0(ArC), 121.7(ArC), 116.4 (ArC), 100.9(=CH), 29.2(CH_3), 18.8(CH), 12.1(CH_2), 12.0 (CH_2); HRMS (ESI-TOF) (*m/z*) $[M + H]^+$ calcd for $C_{13}H_{13}N_2O_2$ 229.0972, found 229.0970.

*3-Cyclopropyl-7-phenylamino-isoquinoline-5,8-dione (**7o**)*

Red solid; yield 64%; mp 133–135 °C; ^{1}H NMR ($CDCl_3$, 400 MHz): $\delta = 9.17$ (s, 1H, ArH), 7.74 (s, 1H, ArH), 7.29 (d, $J = 8.0$ Hz, 2H, ArH), 6.79 (d, $J = 8.0$ Hz, 1H, ArH), 6.73–6.71 (m, 2H, ArH), 6.46(s, 1H, ArH), 2.18 (m, 1H, –CH–), 1.29–1.25(m, 4H, 2 × –CH_2–); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 182.8$(C=O), 181.1(C=O), 172.2(N–C), 148.3(ArC), 146.4(ArC), 129.8(ArC), 129.3(ArC), 129.1 (ArC), 126.2(ArC), 122.9(ArC), 118.6(ArC), 116.2(ArC), 115.1(ArC), 115.0(ArC), 103.4(=CH), 18.9(CH), 12.3 (CH_2), 12.1(CH_2); HRMS (ESI-TOF) (*m/z*) $[M + H]^+$ calcd for $C_{18}H_{14}N_2O_2$ 291.1128, found 291.1125.

*3-Cyclopropyl-7-morpholin-4-yl-isoquinoline-5,8-dione (**7p**)*

Red solid; yield 73%; mp 166–168 °C; ^{1}H NMR ($CDCl_3$, 400 MHz): $\delta = 9.04$ (s, 1H, ArH), 7.65 (s, 1H, ArH), 6.05(s, 1H, ArH), 3.89–3.87 (m, 4H, 2 × –CH_2–), 3.60–3.58 (m, 4H, 2 × –CH_2–), 2.24–2.22 (m, 1H, –CH–), 1.28–1.24(m, 4H, 2 × –CH_2–); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 182.5$ (C=O), 182.1(C=O), 170.9(N–C), 153.4(ArC), 148.6 (ArC), 137.4(ArC), 123.7(ArC), 115.4(ArC), 111.2(=CH), 66.4(CH_2), 66.2(CH_2), 49.2(CH_2), 49.1(CH_2), 18.6(CH), 11.9(CH_2), 11.7(CH_2); HRMS (ESI-TOF) (*m/z*) $[M + H]^+$ calcd for $C_{16}H_{16}N_2O_3$ 284.1234, found 285.1232.

*3-Phenyl-7-p-tolylsulfanyl-isoquinoline-5,8-dione (**7q**)*

Red solid; yield 68%; mp 166–168 °C; ^{1}H NMR ($CDCl_3$, 400 MHz): $\delta = 9.23$ (s, 1H, ArH), 8.18 (s, 1H, ArH), 8.09 (d, $J = 8.0$ Hz, 2H, ArH), 7.49–7.48 (m, 3H, ArH), 7.35–7.33 (m, 3H, ArH), 7.14 (d, $J = 4.0$ Hz, 2H, ArH), 2.35 (s, 3H, –CH_3); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta =$ 178.1(C=O), 177.8(C=O), 162.6(N–C), 149.5(ArC), 149.3 (ArC), 147.3(ArC), 138.8(ArC), 138.5(ArC), 138.4(ArC), 137.5(ArC), 131.8(ArC), 131.7(ArC), 130.8(ArC), 130.1 (ArC), 130.0(ArC), 129.8(ArC), 129.0(ArC), 128.5(ArC), 127.5(ArC), 124.2(ArC), 115.5(=CH), 21.3(CH_3); HRMS (ESI-TOF) (*m/z*) $[M + H]^+$ calcd for $C_2H_{15}NO_2S$ 357.0823, found 357.0816.

*3-(4-Methoxy-phenyl)-7-p-tolylsulfanyl-isoquinoline-5,8-dione (**7r**)*

Red solid; yield 72%; mp 171–173 °C; ^{1}H NMR ($CDCl_3$, 400 MHz): $\delta = 9.20$ (s, 1H, ArH), 8.13 (s, 1H, ArH), 8.08 (d, $J = 8.0$ Hz, 2H, ArH), 7.35–7.33 (m, 3H, ArH), 7.17 (d, $J = 4.0$ Hz, 2H, ArH), 7.03 (d, $J = 4.0$ Hz, 2H, ArH), 3.90 (s, 3H, –OCH_3), 2.37 (s, 3H, –CH_3); ^{13}C NMR ($CDCl_3$, 100 MHz): $\delta = 179.3$(C=O), 178.2(C=O), 165.8(N–C), 151.2(ArC), 148.8(ArC), 147.3(ArC), 137.2(ArC), 136.5 (ArC), 136.3(ArC), 133.5(ArC), 131.9(ArC), 131.5 (ArC), 130.1(ArC), 130.0(ArC), 129.8(ArC), 129.6(ArC), 128.5(ArC), 128.3(ArC), 127.1(ArC), 122.5(ArC), 116.3 (=CH), 55.5(OCH_3), 22.5(CH_3); HRMS (ESI-TOF) (*m/z*) $[M + H]^+$ calcd for $C_{23}H_{17}NO_2S$ 387.0929, found 387.0934.

Biological evaluation

Cell viability

Human hepatocellular carcinoma (HepG2), human non-small-cell lung tumor (A549), human colon carcinoma (HCT-116), and human cervical carcinoma (HeLa) were obtained from the Cell Bank of the Chinese Academy of Sciences (Shanghai, China). All cell lines were cultured in a 37 °C incubator with a 5% CO_2 environment. Compounds were dissolved in Dimethyl sulfoxide (DMSO) with a concentration of 10 mM. The final DMSO concentration in the cell culture and cell viability tests is 0.05%. Cells were seeded into 96-well plates at a concentration of 2×10^3 per well, cultured for 24 h, exposed to the compounds at various concentrations for cancer cell viability experiments, cells were cultured for 72 h and viability was determined through the use of the MTT assay.

Fluorescence morphological examination

Hoechst 33342 staining was carried out according to the procedure of literature (Ding et al. 2009). Cells were treated with compound **7a** in 0, 0.1, 5, and 10 μM for 48 h. Then stained by incubating in PBS containing Hoechst 33342 (10 μg/mL) at 37 °C for 3 min in the dark. Then washed cells with PBS three times. Stained cells were viewed under a fluorescence microscope (Leica, German) with 200 magnification.

Conclusion

To find novel cytotoxic anticancer agents, we have prepared eighteen isoquinoline quinones as natural Mansouramycins analogs. Most of these compounds displayed cytotoxicities against MCF-7, A549, HCT116, and HepG2 cancer cell lines at micromolar concentration. Apoptosis of HepG2 cells induced by compound **7a** was observed by Hoechst33342 staining assay. The current study contributes to understand the SAR and mechanisms of anticancer activity of isoquinoline quinones, which would be helpful to develop novel marine derived anticancer agents.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no competing interests.

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