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Synthesis and antitumor activity of 5-[1-(3-(dimethylamino)propyl)-5-halogenated-2-oxoindolin-(3*Z*)-ylidenemethyl]-2,4-dimethyl-1*H*-pyrrole-3-carboxamides

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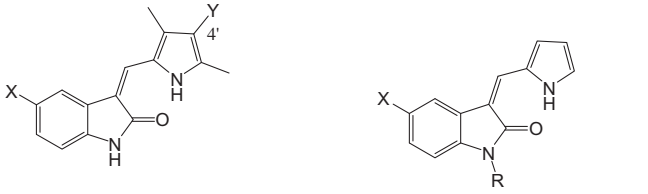
ABSTRACT

We report herein the design and synthesis of novel 1-[3-(dimethylamino)propyl]indolin-2-one derivatives based on the structural features of Sunitinib, a known multitargeted receptor tyrosine kinase inhibitor, and TMP-20, a previously discovered compound with good antitumor activity in our lab. These newly synthesized derivatives were evaluated for in vitro activity against five human cancer cell lines and VEGF/bFGF-stimulated HUVECs. Results revealed that all of the target compounds **1a–p** show potent antitumor activity, compounds **1e–h** (IC_{50}'s: 0.45–5.08 μM) are more active than Sunitinib (IC_{50}'s: 1.35–6.61 μM), and the most active compound **1h** (IC_{50}: 0.47–3.11 μM) is 2.1–4.6-fold more potent than Sunitinib against all five cancer cell lines. In addition, like Sunitinib, **1a–p** have higher selectivity on VEGF-stimulated HUVEC other than bFGF-stimulated HUVEC.

Indolin-2-ones are an important class of compounds with potent antitumor activity, and structural modifications mainly at the 3- and 5-positions of the indolin-2-one ring have recently led many derivatives possessing more potent inhibition on different receptors.[1,2] Semaxanib (SU5416, Fig. 1), the first synthetic indolin-2-one small-molecule compound showed potent activity against vascular endothelial growth factor (VEGF) receptor-1 and -2 tyrosine kinases,[3,4] but the development of Semaxanib was ceased due to the severe toxicity and negative results in Phase II/III studies.[5,6] Sun et al. synthesized a series of novel derivatives by introduction halogens (F, Cl or Br) and basic side chains at the C-5 position of the indolin-2-one ring and C-4′position of the pyrrole ring of SU5416 respectively, and found Sunitinib (Su11248, Fig. 1) to be a new multitargeted receptor tyrosine kinase inhibitor[7] which has activity against many human cancer cell lines.[8–12] The US Food and Drug Administration (FDA) approved Sunitinib for the treatment of advanced renal cancer and gastrointestinal stromal tumor (GIST) after disease progression on or intolerance to Imatinib mesylate in January 2006.[13]

Z24 (Fig. 1) with a piperidin-1-ylmethyl group at the N-1 position of indolin-2-one ring was designed and synthesized by our lab. It can inhibit angiogenesis in new blood vessels, resulting in growth suppression of various types of tumors in vivo.[14–16] TMP-20 (Fig. 1), a novel derivative with a 3-(dimethylamino)propyl group and a chlorine atom at the N-1 and C-5 positions of indolin-2-one ring respectively, was also discovered in our lab. Compared with Z24 (a Mannich base), TMP-20 has more chemical stability while retaining the good antitumor activity of Z24 (not published).



Semaxanib (X = Y = H)
Sunitinib (X = F, Y = $Et_2N(CH_2)_2NHCO$)
Z24 (X = H, R = N-CH_2)
TMP-20 (X = Cl, R = $Me_2N(CH_2)_3$)

Figure 1. Structures of some indolin-2-ones.

As part of our continuing investigation of indolin-2-ones as potential antitumor drug candidates, we planned to design and synthesize a series of novel indolin-2-one derivatives. This design combined the structural features of Sunitinib and TMP-20: a 3-(dimethylamino)propyl group and a halogen atom (F or Cl) at the N-1 and C-5 positions of indolin-2-one ring respectively, and diversified substituted carbamoyl groups at the C-4′position of the pyr-

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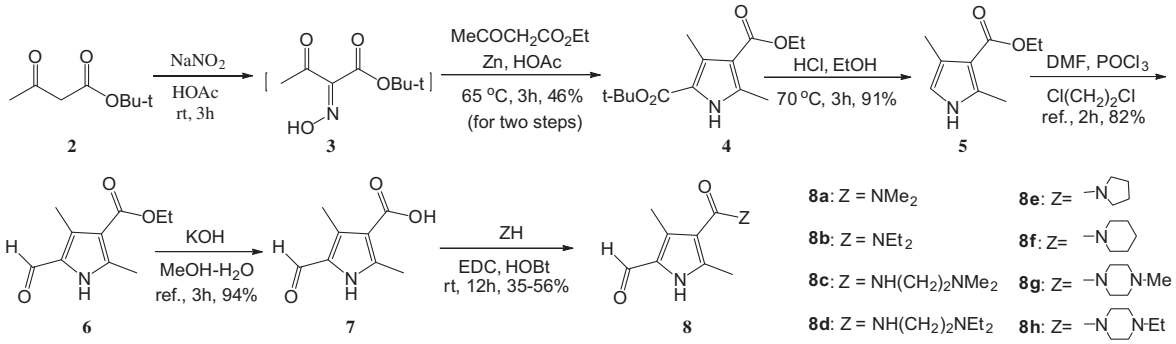
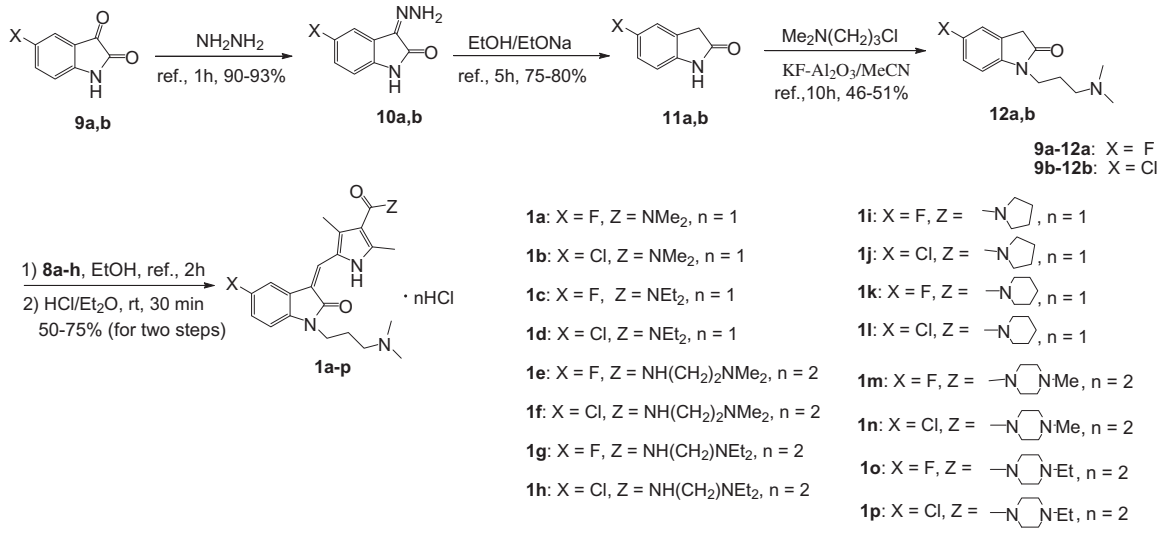


Scheme 1. Synthesis of pyrrole-4-carboxamides **8a–h**.



Scheme 2. Synthesis of novel indolin-2-ones **1a–p**.

role ring. Our primary objective was to optimize the potency of these compounds against human cancer cell lines. A structure–activity relationship (SAR) study was also explored to facilitate the further development of the indolin-2-one derivatives.

Detailed synthetic pathways to pyrrole-3-carboxamides **8a–h** and indolin-2-ones **1a–p** are depicted in Schemes 1 and 2, respectively. According to well-established literature procedures,[7] condensation of commercially available *tert*-butyl acetoacetate (**2**) with sodium nitrite in acetic acid gave oxime **3**, and subsequently reductive cyclization with ethyl acetoacetate yielded bis-ester **4**. Elective hydrolytic decarboxylation of **4** and then formylation afforded aldehyde **6**. Base hydrolysis of the ethyl ester of **6** provided the key intermediate 5-formyl-2, 4-dimethylpyrrole-3-carboxylic acid **7**, which upon amidation with different amines (ZH) in the presence of 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride (EDC) and *N*-hydroxybenzotriazole (HOBt) was converted to pyrrole-3-carboxamides **8a–h** (Scheme 1).

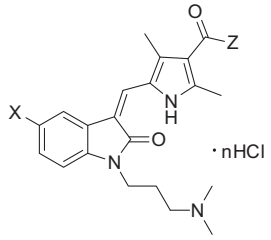
Reduction of the ketone moiety of isatins **9a,b** via Wolff–Kishner–Huang reaction gave indolin-2-ones **11a,b** according to literature procedures,[17] which upon nucleophilic substitution with 1-chloro-3-dimethylaminopropane hydrochloride in the presence of KF-Al_2O_3 in refluxing acetonitrile was converted to 1-(3-dimethylaminopropyl)indolin-2-ones **12a,b**. Finally, the target compounds **1a–p** were prepared via aldol condensation of **12a,b** with pyrrole-3-carboxamides **8a–h**, and further hydrochlorate formation with hydrogen chloride gas in diethyl ether (Scheme 2). All of the new synthesitic compounds were well characterized through the spectral characteristics.[18] As expected, the pyrrole-2-methylidene geometry at the 3-position of indolin-2-one ring was confirmed to have the Z-configuration by NOE NMR experiments.

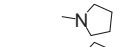
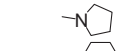
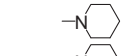
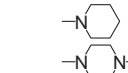
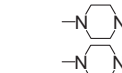
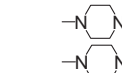
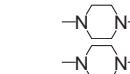
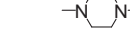
All the newly synthesized indolin-2-ones **1a–p** were evaluated for in vitro antitumor activity in five human cancer cell lines, including IM-9 (B-lymphoblastoid), A549 (lung adenocarcinoma), HL-60 (promyelocytic leukemia), K-562 (erythromyeloblastoid) and MDA-MB-231 (breast cancer) using standard MTT assay.[19] The 50% inhibition concentrations (IC_{50}'s) were compared with those of Sunitinib (Table 1).

The indolin-2-ones **1a–p** have potent in vitro antitumor activity against the tested human cancer cell lines. Among them, compouds **1e–h**, **1n** and **1p** (IC_{50}'s: 0.45–8.88 μM) are more potent than or comparable to Sunitinib (IC_{50}'s: 1.35–6.61 μM) against all of the cell lines. In particular, compound **1h** (IC_{50}s: 0.47–3.11 μM) was found to be 2.1–4.6-fold more potent than the reference drug against these cancer cell lines.

According to the biological evaluation results, antitumor activity of the indolin-2-ones **1a–p** depends mainly on the side chains at the C-4′position of the pyrrole ring. The relative contribution of the Z moieties in linear side chains at the same position to antitumor activity is as follows: $NH(CH_2)_2NNEt_2 \approx NH(CH_2)_2NMe_2$ >> $-NEt_2 > NMe_2$, and the antitumor activity of the Z moieties in cyclic side chains are in the order: 4-methyl-1-piperidinyl > 4-ethyl-1-piperidinyl, 1-piperidinyl > 1-pyrrolidinyl. In addition, 5-chloric indolin-2-ones are generally more active than the corresponding 5-fluoric analogs.

Table 1
In vitro activity of the target compounds **1a**–**p** against selected cells



Cell lines				IC_{50}[a] (μM)						
Compounds	X	Z	*n*	IM-9	A549	HL-60	K-562	MDA-MB-231	HUVEC	
									bFGF	VEGF
1a	F	$-NMe_2$	1	17.95	25.19	14.83	13.60	19.22	19.85	5.23
1b	Cl	$-NMe_2$	1	9.42	16.01	7.30	6.74	12.03	8.27	1.62
1c	F	$-NEt_2$	1	14.12	29.89	10.66	4.88	13.74	10.45	3.96
1d	Cl	$-NEt_2$	1	6.28	13.09	4.02	2.07	6.82	4.87	2.90
1e	F	$-NH(CH_2)_2NMe_2$	2	1.36	2.94	2.73	1.46	2.14	1.42	0.48
1f	Cl	$-NH(CH_2)_2NMe_2$	2	0.45	3.52	1.17	0.53	2.44	3.36	1.11
1g	F	$-NH(CH_2)_2NEt_2$	2	1.63	5.08	1.82	1.33	2.86	1.68	0.71
1h	Cl	$-NH(CH_2)_2NEt_2$	2	0.47	3.11	1.07	0.52	2.29	3.08	2.72
1i	F		1	19.05	21.23	11.92	11.60	13.31	11.26	6.56
1j	Cl		1	9.36	14.03	7.82	6.58	11.46	4.65	3.92
1k	F		1	4.83	18.71	4.39	4.09	11.49	5.27	4.06
1l	Cl		1	4.49	7.33	3.99	2.14	5.68	2.04	1.31
1m	F		2	4.06	11.07	7.59	5.27	9.00	5.64	2.99
1n	Cl		2	1.47	6.78	3.56	1.41	5.06	2.00	1.34
1o	F		2	7.26	13.73	13.44	5.98	8.09	3.38	1.58
1p	Cl		2	2.25	8.88	2.62	2.45	3.46	1.77	1.02
Sunitinib				1.35	6.61	3.53	2.41	4.51	4.04	2.75

[a] IC_{50} values were determined by at least three separate tests and reported as mean values.

All compounds were also further examined for inhibitory activity against VEGF- or recombinant human basic fibroblast growth factor (bFGF)- stimulated human umbilical venous endothelial cells (HUVEC) proliferation using standard MTT assay[19] and the results are reported in Table 1. Results suggested that the indolin-2-ones **1a–p** have higher selectivity on VEGF-stimulated HUVEC (IC_{50}'s: 0.48–6.56 μM) other than bFGF-stimulated HUVEC (IC_{50}'s: 1.42–19.85 μM), which is consistent with Sunitinib. Further studies on related mechanism of action and structural modifications are currently in progress.

In summary, we report herein the design and synthesis of some novel 5-[1-(3-(dimethylamino)propyl)-5-halogenated-2-oxoindolin-(3*Z*)-ylidenemethyl]-2,4-dimethyl-1*H*-pyrrole-3-carboxamides based on the structural features of Sunitnib and TMP-20. The newly synthesized compounds were evaluated for their in vitro activity against five human cancer cell lines and VEGF/bFGF-stimulated HUVECs. Our results revealed that all of the target compounds show potent antitumor activity, and the most active compound **1h** is 2.1–4.6-fold more potent than Sunitinib against the tested five cancer cell lines. In addition, **1a–p** have higher selectivity on VEGF-stimulated HUVEC other than bFGF-stimulated HUVEC.

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18. To a stirring solution of **9a** (4.95 g, 30 mmol) in methanol (80 mL) was added dropwise 85% hydrazine hydrate (3.53 mL, 60 mmol) over a period of 30 min at

room temperature. The reaction mixture was heated to refluxing and stirred for 1 h, and then cooled to room temperature and filtrated. The solid obtained was washed with methanol and dried in vacuo to give **10a** (4.85 g, 90.2%) as a yellow solid, mp: 183–185 °C (183–185 °C[20]). A mixture of **10a** (3.58 g, 20 mmol) and sodium ethoxide (6.80 g, 100 mmol) in absolute ethanol (100 mL) was heated to refluxing and stirred for 5 h, and then cooled to room temperature. To the mixture was added slowly ice-water (100 mL), adjusted to pH 1 with 4 N HCl and filtrated. The solid obtained was recrystallized from water to afford **11a** (2.43 g, 80.4%) as an off-white solid, mp: 135–137 °C (135–137 °C[20]). A suspension of KF (23.24 g, 400 mmol) and Al_2O_3 (40.80 g, 400 mmol) in water was stirred for 0.5 h at room temperature, and then concentrated under reduced pressure, dried in vacuo to prepare KF-Al_2O_3(64.04 g, 100%). A mixture of **11a** (6.04 g, 40 mmol), 1-chloro-3-dimethylaminopropane hydrochloride (5.53 g, 35 mmol) and KF-Al_2O_3 (19.21 g, 120 mmol) in anhydrous acetonitrile (150 mL) was heated to refluxing and stirred for 10 h, and then cooled to room temperature and filtered. The filtrate was concentrated under reduced pressure. The crude product was purified by column chromatography (silica gel) eluted with petroleum ether and ethyl acetate (v:v = 1:1) to afford **12a** (4.73 g, 50.1%) as a brown oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 1.85–1.80 (m, 2H), 2.22 (s, 6H), 2.33 (t, 2H, J = 7 Hz), 3.52 (s, 2H), 3.76 (t, 2H, J = 7 Hz), 6.84–6.81 (m, 1H), 7.01–6.94 (m, 2H). A mixture of **12a** (0.73 g, 3.1 mmol), **8a** (0.58 g, 3 mmol) and pyrrolidine (0.3 mL) in methanol (15 mL) was heated to refluxing and stirred for 2 h, and then concentrated under reduced pressure. The residue was purified by column chromatography (silica gel) eluted with methanol and dichloromethane (v:v = 1:40) to yield the free base of **1a**, which was dissolved in absolute diethyl ether, and then pumped dried hydrochloride gas at 0–5 °C for 30 min. The reaction mixture was allowed to stir for another 30 min at room temperature, the resulting solid was collected by suction, and dried in vacuo to give the title compound **1a** (0.62 g, 46.3%, for two steps) as a yellow solid, mp: 120–122 °C. ^{1}H NMR (400 Hz, DMSO − d_6 + D_2O) δ ppm: 2.02–2.05 (m, 2H), 2.27 (s, 3H), 2.30 (s, 3H), 2.77 (s, 6H),2.92–2.99 (m, 6H), 3.11–3.16 (m, 2H), 3.91 (t, 2H, J = 7 Hz), 7.05–7.14 (m, 2H), 7.73–7.77 (m, 2H), 13.46 (s, 1H). FAB-MS: m/z 413.2 $(M+H)^+$.

The other target compounds **1b**–**1p** (yields: 35.1–64.2%) were obtained as yellow solids in a similar manner as for the preparation of **1a**. Compound **1b**, mp: 112–114 °C. ^{1}H NMR (400 Hz, DMSO − d_6 + D_2O) δ ppm: 2.02–2.06 (m, 2H), 2.28 (s, 3H), 2.30 (s, 3H), 2.77 (s, 6H), 2.93–3.11 (m, 6H), 3.13-3.15 (m, 2H), 3.92 (t, 2H, J = 7 Hz), 7.19–7.24 (m, 2H), 7.77 (s, 1H), 7.99 (d, 1H, J = 2 Hz), 13.43 (s, 1H). FAB-MS: m/z 429.1 $(M+H)^+$. Compound **1c**, mp: 178–180 °C. ^{1}H NMR (400 Hz, DMSO − d_6 + D_2O) δ ppm: 1.05 (t, 3H, J = 7 Hz), 1.22 (t, 3H, J = 7 Hz), 2.02–2.10 (m, 2H), 2.22 (s, 3H), 2.31 (s, 3H), 2.82 (s, 6H), 3.15 (t, 2H, J = 8 Hz), 3.30–3.32 (m, 2H), 3.53–3.55 (m, 2H), 3.84 (t, 2H, J = 7 Hz), 7.01–7.03 (m, 2H), 7.42–7.45 (m, 2H), 13.09 (s, 1H). FAB-MS: m/z 441.2 $(M+H)^+$. Compound **1d**, mp: 96–98 °C. ^{1}H NMR (400 Hz, DMSO − d_6) δ ppm: 1.01–1.12 (m, 6H), 2.03–2.07 (m, 2H), 2.26 (s, 3H), 2.28 (s, 3H), 2.71 (s, 3H), 2.76 (s, 3H), 3.08–3.13 (m, 2H), 3.26–3.52 (m, 4H), 3.93 (t, 2H, J = 7 Hz), 7.19–7.23 (m, 2H), 7.81 (s, 1H), 8.05 (s, 1H), 10.53 (brs, 1H), 13.45 (s, 1H). FAB-MS: m/z 458.1 $(M+H)^+$. Compound **1e**, mp: 90–92 °C. ^{1}H NMR (400 Hz, DMSO − d_6 + D_2O) δ ppm: 2.04–2.08 (m, 2H), 2.43 (s, 3H), 2.48 (s, 3H), 2.80 (s, 6H), 2.90 (s, 6H), 3.13–3.17 (m, 2H), 3.30 (t, 2H, J = 6 Hz), 3.66 (t, 2H, J = 6 Hz), 3.91 (t, 2H, J = 7 Hz), 7.06–7.11 (m, 2H), 7.63–7.65 (m, 2H), 13.40 (s, 1H). FAB-MS: m/z 456.2 $(M+H)^+$. Compound **1f**, mp: 200–202 °C. ^{1}H NMR (400 Hz, DMSO − d_6) δ ppm: 2.04–2.07 (m, 2H), 2.49 (s, 6H), 2.70 (s, 3H), 2.71 (s, 3H), 2.81 (s, 3H), 2.82 (s, 3H), 3.10–3.12 (m, 2H), 3.23–3.25 (m, 2H), 3.61–3.63 (m, 2H), 3.93 (t, 2H, J = 7 Hz), 7.21–7.26 (m, 2H), 7.84 (s, 1H), 8.00–8.03 (m, 1H), 8.07–8.09 (m, 1H), 10.67 (br s, 2H), 13.57 (s, 1H). FAB-MS: m/z 472.2 $(M+H)^+$. Compound **1g**, mp: 70–71 °C. ^{1}H NMR (400 Hz, CD_3OD) δ ppm: 1.37 (t, 6H, J = 7 Hz), 2.13–2.16 (m, 2H), 2.47 (s, 3H), 2.51 (s, 3H), 2.87 (s, 6H), 3.20 (t, 2H, J = 8 Hz), 3.28–3.39 (m, 6H), 3.73 (t, 2H, J = 6 Hz), 3.96 (t, 2H, J = 6 Hz), 6.91–6.96 (m, 1H), 7.03–7.08 (m, 1H), 7.47–7.49 (m, 1H), 7.59 (s, 1H), 13.61 (s, 1H). FAB-MS: m/z 484.1 $(M+H)^+$. Compound **1h**, mp: 124–126 °C. ^{1}H NMR (400 Hz, DMSO − d_6 + D_2O) δ ppm: 1.06 (t, 6H, J = 7 Hz), 2.23 (s, 6H), 1.86–1.91 (m, 2H), 2.33 (t, 2H, J = 7 Hz), 2.51 (s, 3H), 2.58–2.64 (m, 7H), 2.70 (t, 2H, J = 6 Hz), 3.50 (q, 2H, J = 5 Hz), 3.88 (t, 2H, J = 7 Hz), 6.59 (br s, 1H), 6.88 (d, 1H, J = 8 Hz), 7.14 (dd, 1H, J_1 = 2 Hz, J_2 = 8 Hz), 7.35 (s, 1H), 7.43 (d, 1H, J = 2 Hz), 13.55 (s, 1H). FAB-MS: m/z 500.1 $(M+H)^+$. **1i**, mp: 138–139 °C. ^{1}H NMR (400 Hz, DMSO − d_6) δ ppm: 1.80–1.89 (m, 4H), 2.00–2.08 (m, 2H), 2.29 (s, 3H), 2.32 (s, 3H), 2.72 (s, 3H), 2.73 (s, 3H), 3.08–3.21 (m, 2H), 3.21–3.24 (m, 2H), 3.44–3.47 (m, 2H), 3.93 (t, 2H, J = 7 Hz), 7.02–7.07 (m, 1H), 7.16–7.20 (m, 1H), 7.77 (s, 1H), 7.84–7.87 (m, 1H), 10.30 (br s, 1H), 13.49 (s, 1H). EI-MS: m/z 438.2 M^+. Compound **1j**, mp: 136–138 °C. ^{1}H NMR (400 Hz, DMSO − d_6) δ ppm: 1.86–1.93 (m, 4H), 2.02–2.08 (m, 2H), 2.30 (s, 3H), 2.32 (s, 3H), 2.72 (s, 3H), 2.73 (s, 3H), 3.08–3.13 (m, 2H), 3.19-3.22 (m, 2H), 3.43–3.46 (m, 2H), 3.93 (t, 2H, J = 7 Hz), 7.19–7.26 (m, 2H), 7.82 (s, 1H), 8.06 (d, 1H, J = 2 Hz), 10.28 (br s, 1H), 13.44 (s, 1H). EI-MS: m/z 454.1 M^+. Compound **1k**, dec.: 263 °C. ^{1}H NMR (400 Hz, DMSO − d_6) δ ppm: 1.48–1.53 (m, 6H), 2.01–2.06 (m, 2H), 2.27 (s, 3H), 2.30 (s, 3H), 2.72 (s, 3H), 2.73 (s, 3H), 3.09–3.14 (m, 2H), 3.43–3.52 (m, 4H), 3.93 (t, 2H, J = 7 Hz), 7.01–7.02 (m, 1H), 7.16–7.19 (m, 1H), 7.77 (s, 1H), 7.83–7.86 (m, 1H), 10.24 (br s, 1H), 13.51 (s, 1H). EI-MS: m/z 452.1 M^+. Compound **1l**, dec.: 256 °C. Yield: 39.3%. ^{1}H NMR (400 Hz, DMSO − d_6) δ ppm: 1.48–1.61 (m, 6H), 1.99–2.07 (m, 2H), 2.26–2.32 (m, 6H), 2.73 (s, 3H), 2.74 (s, 3H),3.11–3.13 (m, 2H), 3.41–3.53 (m, 4H), 3.93 (t, 2H, J = 7 Hz), 7.18–7.26 (m, 2H), 7.82 (s, 1H), 8.07 (d, 1H, J = 2 Hz), 10.01 (br s, 1H), 13.47 (s, 1H). EI-MS: m/z 468.1 M^+. Compound **1m**, mp: 214–216 °C. ^{1}H NMR (400 Hz, DMSO − d_6) δ ppm: 2.02–2.10 (m, 2H), 2.32–2.35 (m, 6H), 2.70 (s, 3H), 2.72 (s, 3H), 2.76–2.78 (m, 3H), 2.99–3.01 (m, 2H), 3.09–3.14 (m, 2H), 3.37–3.46 (m, 6H), 3.93 (t, 2H, J = 7 Hz), 7.02–7.07 (m, 1H), 7.19–7.22 (m, 1H), 7.79 (s, 1H), 7.85–7.88 (m, 1H), 10.82 (br s, 1H), 11.60 (br s, 1H), 13.57 (s, 1H). FAB-MS: m/z 468.2 $(M+H)^+$. **1n**, dec.: 232 °C. ^{1}H NMR (400 Hz, DMSO − d_6) δ ppm: 2.02–2.10 (m, 2H), 2.32–2.35 (m, 6H), 2.70 (s, 3H), 2.72 (s, 3H), 2.76–2.78 (m, 3H), 2.99–3.01 (m, 2H), 3.09–3.14 (m, 2H), 3.37–3.46 (m, 6H), 3.93 (t, 2H, J = 7 Hz), 7.23–7.26 (m, 2H), 7.85 (s, 1H), 8.09 (d, 1H, J = 2 Hz), 10.59 (br s, 1H), 11.38 (br s, 1H), 13.55 (s, 1H). FAB-MS: m/z 484.1 $(M+H)^+$. **1o**, mp: 199–201 °C. ^{1}H NMR (400 Hz, DMSO − d_6) δ ppm: 1.20 (t, 3H, J = 7 Hz), 2.03–2.09 (m, 2H), 2.32–2.35 (m, 6H), 2.71 (s, 3H), 2.73 (s, 3H), 2.92-2.95 (m, 2H), 3.10–3.15 (m, 4H), 3.41–3.56 (m, 6H), 3.93 (t, 2H, J = 7 Hz), 7.03–7.08 (m, 1H), 7.18–7.21 (m, 1H), 7.79 (s, 1H), 7.86–7.88 (m, 1H), 10.47 (br s, 1H), 11.27 (br s, 1H), 13.58 (s, 1H). FAB-MS: m/z 482.2 $(M+H)^+$. **1p**, dec.: 222 °C. ^{1}H NMR (400 Hz, DMSO − d_6) δ ppm: 1.27 (t, 3H, J = 7 Hz), 2.02–2.07 (m, 2H), 2.33 (s, 3H), 2.35 (s, 3H), 2.70 (s, 3H), 2.72 (s, 3H), 2.92–2.95 (m, 2H), 3.09–3.13 (m, 4H), 3.40–3.48 (m, 2H), 3.82–3.88 (m, 4H), 3.95 (t, 2H, J = 7 Hz), 7.23–7.25 (m, 2H), 7.84 (s, 1H), 8.09 (d, 1H, J = 2 Hz), 10.66 (br s, 1H), 11.45 (br s, 1H), 13.54 (s, 1H). FAB-MS: m/z 498.2 $(M+H)^+$.

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