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Research Article

Synthesis of *p-O*-Alkyl Salicylanilide Derivatives as Novel EGFR Inhibitors

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Strategy, Management and Health Policy				
Enabling Technology, Genomics, Proteomics	Preclinical Research	Preclinical Development Toxicology, Formulation Drug Delivery, Pharmacokinetics	Clinical Development Phases I-III Regulatory, Quality, Manufacturing	Postmarketing Phase IV

ABSTRACT Epidermal growth factor receptor (EGFR), a validated target for anticancer drugs, plays a critical role in tumorigenesis and tumor development. A series of *p-O*-alkyl salicylanilide derivatives were designed and synthesized as novel EGFR inhibitors using a salicylic acid scaffold. A simulated six-membered ring strategy formed through intramolecular hydrogen bonds was employed to mimic the planar quinazoline of the EGFR antagonist, gefitinib. The derived compounds with hydroxyl at the ortho position were more potent than ones with methoxyl group. In particular, compounds **5d** and **5b** displayed significant EGFR inhibitory (IC_{50} values = 0.30 and 0.45 μM, respectively) activity as well as potent antiproliferative activity in A431 and HCT-116 tumor cells. These salicylanilides could be considered as promising lead compounds for developing novel EGFR inhibitors. Drug Dev Res 77 : 37–42, 2016.

Key words: epidermal growth factor receptor inhibitors; salicylic acid; salicylanilide; receptor tyrosine kinase

INTRODUCTION

Receptor tyrosine kinases (RTKs) like epidermal growth factor receptor (EGFR) play essential roles in various signal transduction processes. Numerous studies reported that overexpression of EGFR was related to oncogene-associated activities such as cell proliferation, differentiation, migration, and apoptosis [Ding et al., 2012]. EGFR with a cytoplasmic tyrosine kinase domain, a transmembrane domain, and an extracellular domain that binds EGF, is also overexpressed in various tumors [Wissner et al., 2003]. The interaction between EGF and EGFR results in receptor dimerization, autophosphorylation, and activation. EGFR has been validated as an anticancer drug target with two small-molecule EGFR inhibitors, gefitinib (Iressa) and erlotinib (Tarceva) having been approved by FDA for the treatment of cancer [Yang et al., 2012]. These two drugs are among the first-generation ATP-competitive inhibitors and are 4-anilinoquinazoline derivatives (Fig. 1). These outcomes encouraged the development of promising anticancer agents by targeting EGFR.

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Salicylic acid is a phenolic compound isolated from plants and has antiproliferative and anticancer activities [Mahdi et al., 2006]. Extensive studies have focused on evaluating its therapeutic benefits on a broad array of diseases including hypercholesterolemia, hyperglycerolemia, cardiovascular and inflammatory diseases, viral infections, and so forth [Deng and Chow, 2010; Thun et al., 2012]. Salicylic acid derivatives have been evaluated and identified as anticancer agents [Kashfi, 2009; Dovizio et al., 2012].

In an effort to develop novel and potent EGFR inhibitors, a series of *p*-*O*-alkyl salicylanilide derivatives based on the salicylic acid scaffold were designed and prepared in the present study. Structural diversity was assessed by concentrating on the terminal aniline and salicylic acid scaffold (Fig. 2). EGFR inhibitors with quinazoline-based core scaffold were identified as ATP-competitive inhibitors of wild-type EGFR [Zhou et al., 2009]. The quinazoline scaffold binds to the hinge region of EGFR via hydrogen bonds and plays an essential role in enzyme inhibition [Deng et al., 2006]. To develop novel EGFR inhibitors with high affinity for EGFR, salicylanilide scaffold was introduced to form a pseudo six-membered ring via a hydrogen bond between the OH and C=O groups (Fig. 2). The designed structure was used to mimic the rigid and planar conformation of quinazoline scaffold of reported ATP-competitive inhibitors [Wang et al., 2013]. The intramolecular hydrogen bond was supposed to stabilize the planar conformation as well as improve the rigid property. Various terminal anilines were introduced to perform structure activity relationship studies. To validate the importance of the hydroxyl group of the salicylanilide scaffold, another series of *O*-methyl derivatives has also been prepared and evaluated [Gao et al., 2015].

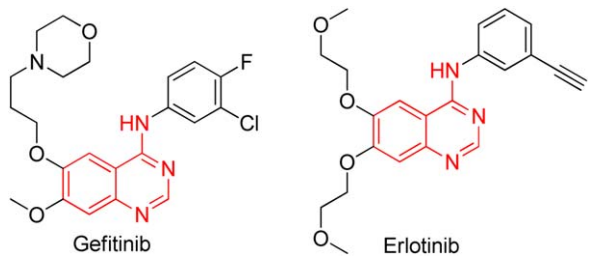


Fig. 1. Structures of approved EGFR inhibitors. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

METHODS AND MATERIALS

Chemistry

General procedure. Reactions were performed under a dry condition realized by standard techniques for the exclusion of moisture. Melting points were determined on electrothermal melting point apparatus and are uncorrected. ^{1}H-NMR spectra are measured with 400 MHz Bruker NMR instrument, with TMS as the internal standard. Electron impact (EI) mass spectra were obtained on a HP-5988A mass spectrometer. The target compounds purification was performed with silica gel column chromatography. Reactions were monitored by thin layer chromatography on silica gel (60GF-254) plates and visualized under a UV lamp.

2-(benzyloxy)-4-methoxybenzaldehyde (**2**) [Bermejo et al., 2002]. To a suspension of 2-hydroxy-4-methoxybenzaldehyde (**1**) (4.56 g, 30 mmol) in dehydrated alcohol (120 mL) was added anhydrous potassium carbonate (12.44 g, 90 mmol) and benzyl chloride (5.20 mL, 45 mmol). The mixture was refluxed for 5 h. Filtration and evaporation of alcohol was done in a vacuum. The residue was extracted with EtOAc (2 × 70 mL). The combined organic

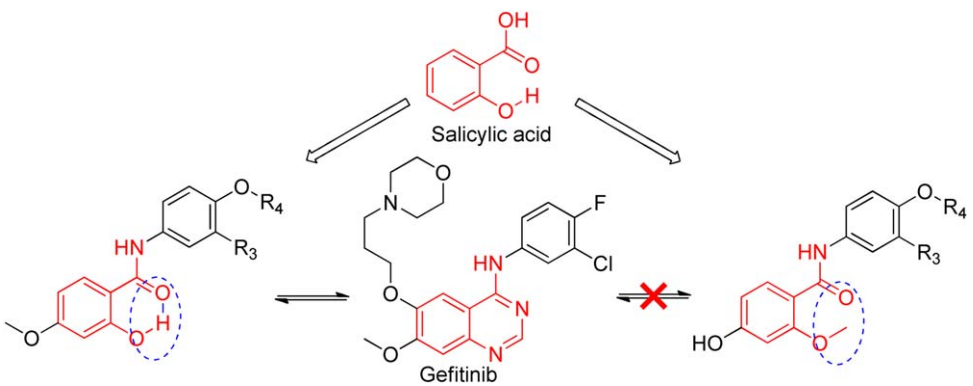


Fig. 2. Design strategy and structures of title compounds based on salicylic acid. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

layers were washed with H_2O (3 × 40 mL), 2 M NaOH (3 × 40 mL), 2 M HCl (3 × 40 mL), and brine (2 × 40 mL), dried over Na_2SO_4, and concentrated to give (**2**) (6.90 g, 95%) as a yellow solid, which was used for the next step without further purification.

4-(benzyloxy)-2-methoxybenzaldehyde (**7**) was prepared using the same procedure described above.

2-(benzyloxy)-4-methoxybenzoic acid (**3**) [Miura et al., 2006]. To a solution of (**2**) (7.27 g, 30 mmol) in THF (100 mL) was added distilled H_2O (40 mL) and NaH_2PO_4 (2.16 g, 18 mmol). The mixture was stirred at room temperature for 20 min. $NaClO_2$ (8.96 g, 99 mmol) and 30% H_2O_2 (6.80 mL, 66 mmol) in distilled H_2O (40 mL) were added. The resultant mixture was stirred at room temperature for 3 h. THF was evaporated under vacuum and the residue was extracted with EtOAc (3 × 70 mL). The combined organic layers were washed with (3 × 30 mL) and the product was extracted with 2 M NaOH (5 × 30 mL). The aqueous phase was acidified with concentrated HCl and the solid obtained was collected by filtration and dried to give (**3**) (7.210 g, 93%) as a white solid.

4-(benzyloxy)-2-methoxybenzoic acid (**8**) was prepared using the same procedure described above.

2-(benzyloxy)-4-methoxy-N-(4-(2-morpholinoethoxy) phenyl) benzamide (**4a**) [Nobuaki et al., 2009]. To a solution of compound (**3**) (1.29 g, 5 mmol) in anhydrous THF (25 mL) were added EDCI (1.71 g, 8.92 mmol), HOBt (1.20 g, 8.92 mmol), and triethylamine (2 mL). After stirring at room temperature for 2 h, 4-(2-morpholinoethoxy) aniline (1.78 g, 8mmol) in anhydrous THF (15 mL) was added drop wise and the reaction continued for 48 h at room temperature. Water (300 mL) was then added and the mixture was stirred for 30 min. The water layer was extracted with AcOEt (50 mL × 3). The organic layer was combined and washed with water and brine, and dried over Na_2SO_4. The filtration and concentration in vacuo afforded (**4a**) (0.78 g, 92%) are as white solid.

4-(benzyloxy)-2-methoxy-N-(4-(2-morpholinoethoxy) phenyl) benzamide (**9a**) was prepared using the same procedure described above.

2-hydroxy-4-methoxy-N-(4-(2-morpholinoethoxy) phenyl) benzamide (**5a**) [Bazin et al., 2013]. To a solution of compound (**4a**) (2.31 g, 5 mmol) in 50 mL of anhydrous MeOH was added Pd/C 10% (51 mg, 0.05 mmol). The suspension was refluxed for 5 h. Pd/C was filtered on Celite®, and the filtrate was concentrated in vacuo. The crude product was purified by silica gel chromatography using dichloromethane as eluent to afford the title compound (**5a**) (1.77 g, 95% yield). m.p.: 162–163°C. EI-MS (*m/z*): 372.2 $[M]^+$. ^1H NMR (400 MHz, $CDCl_3$) δ 7.51 (d, $J = 7.8$ Hz, 2H), 7.39 (d, $J = 8.0$ Hz, 1H), 7.24 (s, 1H), 6.91 (d, $J = 7.8$ Hz, 2H), 6.89 (s, 1H), 4.18 (t, $J = 4.2$ Hz, 2H), 3.75–3.78 (m, 4H), 2.83 (t, $J = 4.2$ Hz, 2H), 2.61 (s, 4H).

The title compounds (**5b–5g)** and (**10b–10g**) were also prepared using the general procedure described above.

N-(4-(3-(dimethylamino) propoxy)-3-fluorophenyl)-2-hydroxy-4-methoxybenzamide (**5b**). m.p.: 139–140°C. EI-MS (*m/z*): 362.0 $[M]^+$. ^1H NMR (400 MHz, DMSO-d6) δ 7.44 (d, $J = 8.0$ Hz, 1H), 7.38 (d, $J = 8.0$ Hz, 1H), 7.35 (d, $J = 8.0$ Hz, 2H), 7.14 (d, $J = 8.0$ Hz, 1H), 7.07 (t, $J = 4.0$ Hz, 1H), 6.93 (d, $J = 8.0$ Hz, 2H), 4.01 (t, $J = 4.0$ Hz, 2H), 3.78 (s, 6H), 3.09–3.15 (m, 2H), 2.48 (s, 2H).

N-(3-fluoro-4-(2-morpholinoethoxy)phenyl)-2-hydroxy-4-methoxybenzamide (**5c**). m.p.: 172–173°C. EI-MS (*m/z*): 390.2 $[M]^+$. ^1H NMR (400 MHz, DMSO-d6) δ 7.82 (dd, $J = 8.0$ Hz, 1H), 7.49 (d, $J = 8.0$ Hz, 1H), 7.45 (dd, $J = 8.0$ Hz, 1H), 7.38 (d, $J = 8.0$ Hz, 1H), 7.27 (t, $J = 4.0$ Hz, 1H), 7.01 (d, $J = 4.0$ Hz, 1H), 4.42 (s, 2H), 3.83 (s, 4H), 3.61 (s, 4H), 3.39 (s, 2H).

N-(3-fluoro-4-(3-morpholinopropoxy)phenyl)-3-hydroxy-4-methoxybenzamide (**5d**). m.p.: 169–170°C. EI-MS (*m/z*): 404.3 $[M]^+$. ^1H NMR (400 MHz, DMSO-d6) δ 7.71 (dd, $J = 8.0$ Hz, 1H), 7.50 (t, $J = 4.0$ Hz, 2H), 7.38 (d, $J = 8.0$ Hz, 1H), 7.12 (t, $J = 4.0$ Hz, 1H), 6.99 (d, $J = 8.0$ Hz, 1H), 4.10 (t, $J = 4.0$ Hz, 2H), 3.65 (s, 4H), 2.51–2.57 (m, 3H), 1.95 (s, 1H).

N-(3-chloro-4-(2-morpholinoethoxy)phenyl)-3-hydroxy-4-methoxybenzamide (**5e**). m.p.: 177–178°C. EI-MS (*m/z*): 406.2 $[M]^+$. ^1H NMR (400 MHz, DMSO-d6) δ 7.69 (d, $J = 8.0$ Hz, 2H), 7.45 (dd, $J = 8.0$ Hz, 1H), 7.39 (d, $J = 8.0$ Hz, 1H), 6.99 (s, 1H), 6.95 (d, $J = 8.0$ Hz, 2H), 4.35–4.41 (m, 2H), 3.79 (s, 4H), 3.51 (s, 4H), 3.40 (s, 2H).

N-(3-chloro-4-(2-morpholinoethoxy)phenyl)-3-hydroxy-4-methoxybenzamide (**5f**). m.p.: 168–169°C. EI-MS (*m/z*): 406.1 $[M]^+$. ^1H NMR (400 MHz, DMSO-d6) δ 7.75 (dd, $J = 8.0$ Hz, 1H), 7.49 (d, $J = 8.0$ Hz, 1H), 7.42 (dd, $J = 8.0$ Hz, 1H), 7.37 (d, $J = 8.0$ Hz, 1H), 7.18 (t, $J = 4.0$ Hz, 1H), 6.97 (d, $J = 4.0$ Hz, 1H), 4.37 (s, 2H), 3.90 (s, 4H), 3.68 (s, 4H), 3.40 (s, 2H).

N-(3-chloro-4-(3-morpholinopropoxy)phenyl)-3-hydroxy-4-methoxybenzamide (**5g**). m.p.: 156–157°C. EI-MS (*m/z*): 420.0 $[M]^+$. ^1H NMR (400 MHz, DMSO-d6) δ 7.69 (dd, $J = 8.0$ Hz, 1H), 7.48 (t, $J = 4.0$ Hz, 2H), 7.40 (d, $J = 8.0$ Hz, 1H), 7.11 (t, $J = 4.0$ Hz, 1H), 6.72 (d, $J = 8.0$ Hz, 1H), 4.07 (t, $J = 4.0$ Hz, 2H), 3.70 (s, 4H), 2.57–2.63 (m, 3H), 1.99 (s, 1H).

N-(4-(3-(dimethylamino)propoxy)phenyl)-4-hydroxy-2-methoxybenzamide (**10a**). m.p.: 162–163°C. EI-MS (*m/z*): 344.3 $[M]^+$. ^{1}H NMR (400 MHz, DMSO-d6) δ 7.70 (d, $J = 8.0$ Hz, 2H), 7.61 (s, 1H), 7.49 (d, $J = 8.0$ Hz, 1H), 7.01–7.06 (m, 1H), 6.97 (d, $J = 8.0$ Hz, 2H), 3.98 (s, 2H), 3.02–3.07 (m, 2H), 2.71 (s, 6H), 2.09 (s, 2H).

N-(4-(2-(dimethylamino)ethoxy)phenyl)-4-hydroxy-2-methoxybenzamide (**10b**). m.p.: 197–198°C. EI-MS (*m/z*): 330.2 $[M]^+$. ^{1}H NMR (400 MHz, DMSO-d6) δ 7.63 (d, $J = 8.0$ Hz, 2H), 7.49 (s, 1H), 7.45 (d, $J = 8.0$ Hz, 1H), 7.01 (d, $J = 8.0$ Hz, 2H), 6.90 (d, $J = 8.0$ Hz, 1H), 4.09 (s, 2H), 2.44 (s, 6H), 1.95 (s, 2H).

N-(4-(2-(diethylamino)ethoxy)phenyl)-4-hydroxy-2-methoxybenzamide (**10c**). m.p.: 173–174°C. EI-MS (*m/z*): 358.3 $[M]^+$. ^{1}H NMR (400 MHz, DMSO-d6) δ 7.70 (d, $J = 8.0$ Hz, 2H), 7.57 (d, $J = 8.0$ Hz, 1H), 7.49 (dd, $J = 8.0$ Hz, 1H), 7.01 (d, $J = 8.0$ Hz, 2H), 6.92 (d, $J = 8.0$ Hz, 1H), 4.34 (s, 2H), 3.12 (s, 4H), 2.48 (d, $J = 8.0$ Hz, 2H), 1.20 (t, $J = 4.0$ Hz, 6H).

4-hydroxy-2-methoxy-N-(4-(2-(pyrrolidin-1-yl) ethoxy)phenyl)benzamide (**10d**). m.p.: 183–184°C. EI-MS (*m/z*): 356.4 $[M]^+$. ^{1}H NMR (400 MHz, DMSO-d6) δ 7.71 (dd, $J = 8.0$ Hz, 2H), 7.59 (d, $J = 8.0$ Hz, 1H), 7.51 (dd, $J = 8.0$ Hz, 1H), 6.97–67.00 (m, 2H), 6.87–6.92 (m, 1H), 4.15 (t, $J = 4.0$ Hz, 2H), 2.99 (s, 2H), 2.78 (s, 4H), 1.80 (s, 4H).

4-hydroxy-2-methoxy-N-(4-(2-(piperidin-1-yl) ethoxy)phenyl)benzamide (**10e**). m.p.: 191–192°C. EI-MS (*m/z*): 370.3 $[M]^+$, ^{1}H NMR (400 MHz, DMSO-d6) δ 7.75 (s, 2H), 7.60 (d, $J = 8.0$ Hz, 1H), 7.54 (dd, $J = 8.0$ Hz, 1H), 7.01 (d, $J = 8.0$ Hz, 2H), 6.95 (d, $J = 8.0$ Hz, 1H), 4.32 (t, $J = 4.0$ Hz, 2H), 3.20 (s, 2H), 3.01 (s, 4H), 1.71–1.75 (m, 4H), 1.54 (s, 2H).

4-hydroxy-2-methoxy-N-(4-(2-morpholinoethoxy) phenyl)benzamide (**10f**). m.p.: 152–153°C. EI-MS (*m/z*): 372.2 $[M]^+$. ^{1}H NMR (400 MHz, DMSO-d6) δ 7.71 (d, $J = 8.0$ Hz, 2H), 7.55 (d, $J = 8.0$ Hz, 1H), 7.49 (dd, $J = 8.0$ Hz, 1H), 7.01 (d, $J = 8.0$ Hz, 2H), 6.90 (d, $J = 8.0$ Hz, 1H), 4.31 (s, 2H), 3.88 (s, 2H), 3.79 (s, 4H), 3.21 (s, 4H).

4-hydroxy-2-methoxy-N-(4-(3-morpholinopropoxy) phenyl)benzamide (**10g**). m.p.: 161–162°C. EI-MS (*m/z*): 386.1 $[M]^+$. ^{1}H NMR (400 MHz, DMSO-D6) δ 7.70 (dd, $J = 8.0$ Hz, 2H), 7.56 (d, $J = 8.0$ Hz, 1H), 7.49 (dd, $J = 8.0$ Hz, 1H), 6.94–6.99 (m, 2H), 6.86 (d, $J = 4.0$ Hz, 1H), 3.99 (t, $J = 4.0$ Hz, 2H), 3.78 (s, 4H), 3.01 (s, 6H), 2.04–2.09 (m, 2H).

Biology

Inhibition of EGFR

EGFR tyrosine kinase activity was measured as the percent of ATP consumed following the kinase reaction as described by Xin et al. [2012]. EGFR kinase reactions were initiated by adding test compounds (1 μL/well) and EGFR (4 μL/well) to a 384-well plate. The assay plate was centrifuged with 1000 rpm for 1 min to mix the reagents and then pre-incubated at 30°C for 30 min. ATP was added to the assay plate. The assay plate was incubated at 30°C for 1 h. ADP-Glo Reagent was added to each well (10 μL/well, Promega), after which the assay plate was incubated at 27°C for 40 min. Kinase detection reagent (20 μL/well) was added to each well and the assay plate was incubated at 27°C for 30 min. The luminescence signal was read with Envision (PerkinElmer). IC_{50} values were calculated according to the observed inhibition ratios.

Inhibition of cell proliferation

The antiproliferative activity of the title compounds against A431 and HCT-116 cancer cells were evaluated by 3-(4, 5-dimethylthiazol-2-yl)-2, 5-diphenyltetrazolium bromide (MTT) method [Wang et al., 2014]. The cells of 1×10^4 per well were plated in 96-well culture plates and allowed to adhere. Then, the cells were incubated with indicated concentrations of the compounds for 24 h. After incubation, MTT solution (20 μL of 5 mg/mL) in PBS was added to each well. Incubation was continued until purple precipitates were visible. Then, the medium was carefully removed and the precipitates were dissolved in 150 μL of DMSO. Absorbance values were determined by a microplate reader (Bio-Rad Instruments) at 490 nm. The IC_{50} values were calculated according to the observed inhibition ratios.

RESULTS AND DISCUSSION

Chemistry

A convenient method for the synthesis of *p-O*-alkyl salicylanilide derivatives was described in Figure 3. The target compounds were derived from commercial available 2-hydroxy-4-methoxybenzaldehyde (**1**) and 4-hydroxy-2-methoxybenzaldehyde (**6**) in four steps. Initially, hydroxyl groups of (**1**) and (**6**) were protected by benzyl to afford compounds (**2**) and (**7**), respectively. The aldehyde groups in (**2**) and (**7**) were both oxidized and converted into carboxyl group to provide the key intermediates (**3**) and (**8**). Thereafter, compounds (**3**) and (**8**) were coupled with various *p-O*-alkyl anilines in the presence of HOBt with EDCI as condensing agent to provide the critical intermediates (**4a–4g**) and (**9a–9g**). Finally, benzyl deprotection with palladium/carbon in methanol yielded the target compounds (**5a–5g**) and (**10a–10g**).

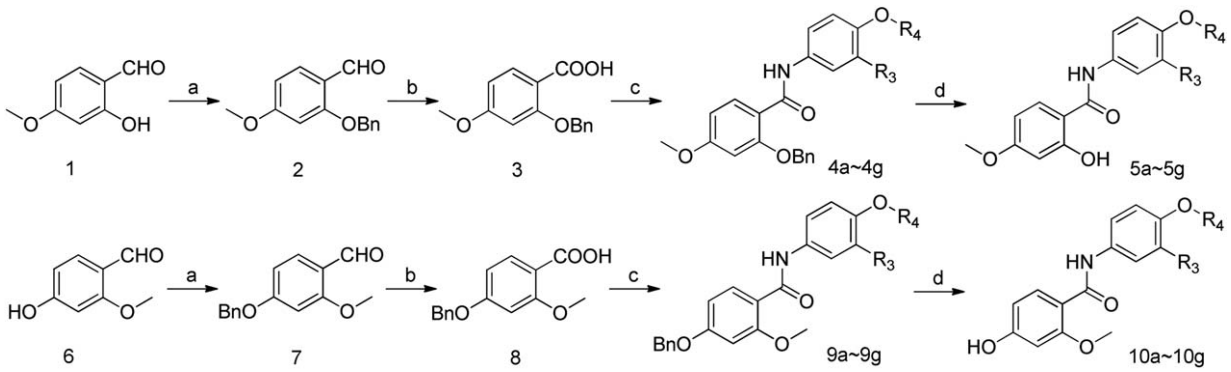
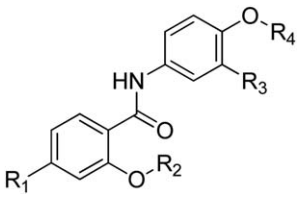


Fig. 3. Synthetic route of the title compounds (**5a–5g**) and (**10a–10g**). *Reagents and conditions*: (**a**) BnCl, K2CO3, Ethanol, reflux; (**b**) H2O2, NaClO2, NaH2PO4; (**c**) EDCI, HOBt, triethylamine, THF; (**d**) Pd/C, H2, Methanol.

TABLE 1. The Structures and Activities of the Title Compounds (5a-5g) and (10a-10g)



	R1	R2	R3	R4	A431 (μM)	HCT-116 (μM)	EGFR (μM)
5a	OMe	H	H		46.25	11.43	4.82
5b	OMe	H	F		10.31	4.38	0.45
5c	OMe	H	F		42.59	46.44	9.06
5d	OMe	H	F		7.08	5.53	0.30
5e	OMe	H	Cl		81.77	143.74	13.29
5f	OMe	H	Cl		4.35	31.28	7.11
5g	OMe	H	Cl		4.85	8.62	6.74
10a	OH	Me	H		97.13	235.18	47.62
10b	OH	Me	H		119.38	698.90	ND
10c	OH	Me	H		102.93	218.92	70.10
10d	OH	Me	H		94.57	54.02	82.00
10e	OH	Me	H		82.18	165.68	97.20
10f	OH	Me	H		77.39	649.16	124.31
10g	OH	Me	H		102.24	16.14	64.47
		Gefitinib			0.98	3.47	0.012

Biological Evaluation

All compounds were evaluated for their EGFR inhibitory activity as well as the antiproliferative potency against A431 and HCT-116 cells using gefitinib as positive control [Mu et al., 2001]. As shown in Table 1, the majority of salicylanilide derivatives displayed EGFR inhibitory activity and antiproliferative potency. Among them, **5b** and **5d** potently inhibited EGFR with IC_{50} values of 0.45 and 0.30 μM, respectively. The IC_{50} values of target compounds (**5a–5g**) ranged from 0.30 μM to 13.29 μM.

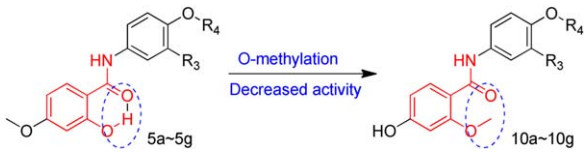


Fig. 4. *O*-methylation decreased the EGFR inhibitory activity. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

Incorporation of the methyl group into the phenolic hydroxyl group (**10a–10g**) resulted in a decrease in potency (Fig. 4). The target compounds (**10a–10g**) with *O*-methyl substituents were much less potent (with mean IC_{50} value of 80.95 μM) than the ones with hydroxyl group (with mean IC_{50} value of 5.97 μM, **5a–5g**). It is assumed that compounds (**10a–10g**) may not be able to form intramolecular hydrogen bond. These results indicated that the intramolecular hydrogen bond may be essential for the EGFR inhibitory activity by forming the pseudo six-membered ring.

All the target compounds were also evaluated for their antiproliferative activity against the cancer cell lines A431 and HCT-116. Their anticancer potency (IC_{50}) is shown in Table 1. The majority of the target compounds inhibited the growth of the tested cancer cell lines in a dose-dependent manner. In inhibiting the growth of HCT-116 cells, compounds with an hydroxyl group (mean IC_{50} value of 35.92 μM) exhibited more potent activities than those

with *O*-methyl substituents (mean IC_{50} value of 291.14 μM). In inhibiting the growth of A431 cells, compounds with an hydroxyl group (with mean IC_{50} value of 28.17 μM) also showed more potent activities than ones with *O*-methyl substituents (mean IC_{50} value of 96.55 μM). Compounds **5b** and **5d** also displayed activity in the antiproliferative assay with IC_{50} values of 10.31 and 4.38 (**5b**), 7.08 and 5.53 (**5d**) μM against A431 and HCT-116 cell lines, respectively.

CONCLUSION

In summary, 14 salicylanilide derivatives were designed and synthesized using a salicylic acid core scaffold as the hinge binding group to inhibit EGFR. These compounds extended the structural diversity of EGFR inhibitors in the development of novel antitumor agents. The improved binding affinity was considered to be contributed by the obvious fit of salicylanilide with EGFR. SAR analysis indicated that the pseudo six-membered ring formed by an intramolecular hydrogen bond played significant roles in EGFR binding affinity and the inhibitory activities.

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