Synthesis and Biological Evaluations of 3-Substituted Indolin-2-ones: A Novel Class of Tyrosine Kinase Inhibitors That Exhibit Selectivity toward Particular Receptor Tyrosine Kinases

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3-Substituted indolin-2-ones have been designed and synthesized as a novel class of tyrosine kinase inhibitors which exhibit selectivity toward different receptor tyrosine kinases (RTKs). These compounds have been evaluated for their relative inhibitory properties against a panel of RTKs in intact cells. By modifying the 3-substituted indolin-2-ones, we have identified compounds which showed selective inhibition of the ligand-dependent autophosphorylation of various RTKs at submicromolar levels in cells. Structure−activity analysis for these compounds and their relative potency and selectivity to inhibit particular RTKs has determined that (1) 3-[(five-membered heteroaryl ring)methylidenyl]indolin-2-ones are highly specific against the VEGF (*Flk-1*) RTK activity, (2) 3-(substituted benzylidenyl)indolin-2-ones containing bulky group(s) in the phenyl ring at the C-3 position of indolin-2-ones showed high selectivity toward the EGF and Her-2 RTKs, and (3) the compound containing an extended side chain at the C-3 position of the indolin-2-one (**16**) exhibited high potency and selectivity when tested against the PDGF and VEGF (*Flk-1*) RTKs. Recent published crystallographic data for two of these 3-substituted indolin-2-ones provides a rationale to suggest that these compounds may bind in the ATP binding pocket of RTKs. The structure−activity analysis supports the use of subsets of these compounds as specific chemical leads for the development of RTK-specific drugs with broad application for the treatment of human diseases.

Introduction

Receptor tyrosine kinases (RTKs) have been shown to be important mediators of cellular signal transduction in cells.[1,2] Many RTKs have been shown to be oncogene products implicating their role in the transformation process associated with human cancers.[3] In this regard, deregulated RTK activity has been shown to play a role in abnormal cell functions associated with the growth, survival, and spread of human tumors.[4] In a broader sense, increased RTK activity has been implicated in atherosclerosis, vascular restenosis, fibrosis of the lung, liver, and kidney, inflammatory disease, and other immune-mediated disorders.[2,3] As a consequence of these associations, RTKs have been viewed as attractive therapeutic targets for the development of novel agents with novel mechanisms to treat human disease.[5] Over the past decade, many investigators have sought to identify small molecules that block RTK function. However, only recently has it been possible to identify several chemical classes that would inhibit kinase activity associated with particular RTK types.[6−8] In this regard, in vitro testing of compounds for inhibitory properties against a wide variety of kinases has provided the rationale to pursue particular chemical subtypes as RTK-specific drug leads.

Recent studies to identify inhibitors of the epidermal growth factor (EGF) RTK have been quite successful. (Phenylamino)quinazolines,[9] pyridopyrimidines,[10] pyrrolopyrimidines,[11] and pyrazolopyrimidines[12] have been identified and exhibit promising in vitro and in vivo potency and specificity toward the EGF RTK. These chemical classes have provided the basis for the selection of drug candidates for the treatment of EGF-dependent human cancers. Biochemical and kinetic studies using the 4-(phenylamino)quinazolines have suggested that these compounds inhibit the EGF RTK in an ATP-competitive manner.[13] Models based upon the cAMP-dependent kinase have been used to help define how these molecules may localize in the ATP binding pocket of the EGF RTK.[11,13] The planar quinazoline core structure has been proposed to bind to the ATP binding cleft and the substituents around the core structure have been suggested to confer specificity through unique interactions between these inhibitors and a few kinase-specific amino acid residues present in the ATP binding sites of the EGF RTK.[13]

Recently, we have elucidated the crystallographic structure of 3-substituted indolin-2-ones in the catalytic core of the fibroblast growth factor (FGF) RTK.[14] In this study, the indolin-2-one core was shown to occupy a site in which the adenine of adenosine triphosphate binds and the substituents around the indolin-2-one core were shown to extend into the hinge region between the two kinase lobes. In this case, compound **42** (3-[(3-(2-carboxyethyl)-4-methylpyrrol-2-yl)methylidenyl]indolin-2-one) (Table 2) was shown to inhibit the FGF RTK in a specific manner and induced conformational changes in the nucleotide-binding loop that may, in part, explain the potency and specificity of this compound. In the present study, we provide data to suggest that various substituents around the indolin-2-one core can contribute to the potency and specificity of this chemical class

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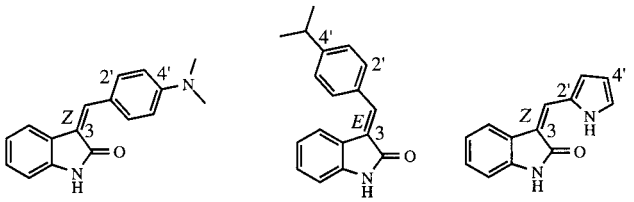
Chart 1. Structures and Biological Evaluations of the Three Lead 3-Substituted Indolin-2-ones (**1**−**3**)



1 **2** **3**

Compound ID	Inhibition of Cellular Tyrosine Kinase Activity (IC_{50}, μM)				
	PDGFR	FLK-1	EGFR	HER-2	IGF-1R
1	19.4	0.8	>100	>100	>100
2	24.2	5.2	18.5	16.9	10.0
3	12.0	0.39	>100	>100	>100

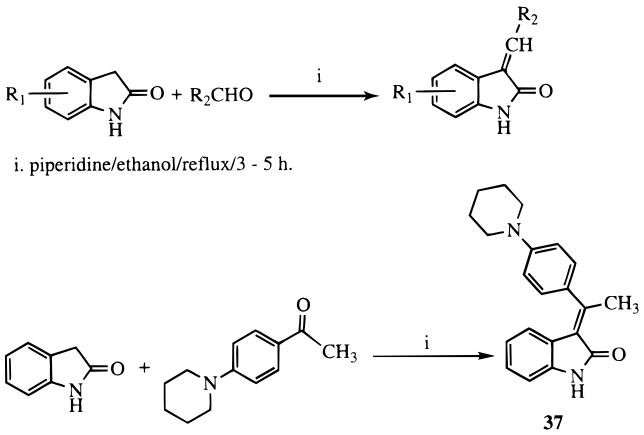
on various RTKs. This study provides the basis to associate subsets of 3-substituted indolin-2-ones as specific chemical leads for the development of particular RTK-specific drugs with broad application to the treatment of human diseases.

Discovery and Design of 3-Substituted Indolin-2-ones as RTK Inhibitors

Using a random-screening approach, we have identified three related compounds of the 3-substituted indolin-2-ones (**1**−**3** in Chart 1) that exhibited inhibitory properties against various RTKs using whole-cell ligand-dependent autophosphorylation assays. As shown in Chart 1, both **1** and **3** were found to be potent and selective inhibitors of the vascular endothelial growth factor (VEGF) [fetal liver kinase-1 (*Flk-1*)] RTKs, whereas **2** was found to be nonselective for RTK inhibition. A comparison of the chemical structures of these compounds provided the foundation to suggest that various structural features may impart changes in RTK specificity. Compound **1** contains a nitrogen atom attached to the C-4′ position of the phenyl at the C-3 position of the indolin-2-one, whereas **2** contains an isosteric methine moiety. In addition, NMR analysis of these compounds indicated differences in *E* and *Z* isomer populations. Both **1** and **3** exist as *Z* isomers, but **2** adopts the *E* isomeric form. This preliminary analysis suggested that the relative potency and selectivity of these compounds for inhibition of these kinases may be sensitive to the substituent at the C-4′ position of **1** or the predominant *Z*−*E* configurations.

To extend these observations, we sought to expand the chemical diversity of this structural type and determine the structure−activity relationship (SAR) for 3-substituted indolin-2-one analogues using a panel of cell-based RTK assays. Five classes of 3-substituted indolin-2-ones were designed and synthesized: (A) compound **1** analogues with different substituent(s) on the phenyl ring at the C-3 position of the indolin-2-one core (Table 1); (B) compound **3** analogues having different substituent(s) in the pyrrole ring at the C-3 position of indolin-2-one (Table 2); (C−E) three classes

Scheme 1. Preparations of 3-Substituted Indolin-2-ones



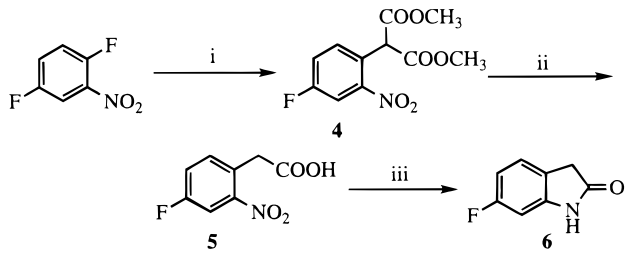
of compounds containing different five-membered heteroaryl rings, including thiophene (Table 3), furan (Table 4), and pyrazole (Table 5), at the C-3 position of indolin-2-one. The impact of the substituents on the indolin-2-one ring on the potency and selectivity of the inhibitors was also examined.

Chemistry

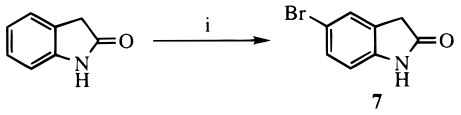
3-Substituted indolin-2-ones were prepared by condensing substituted indolin-2-ones and aldehydes or ketones in the presence of bases (Scheme 1). Indolin-2-one (oxindole), 5-chlorooxindole, and most of the aldehydes were commercially available, whereas 5-nitro-, 4-methyl-, 5-bromo-, 5-methyl-, and 6-fluorooxindoles and a few aldehydes have been prepared by means of reported or newly developed methods. 5-Nitrooxindole was prepared by nitration of the oxindole at −5 to 0 °C followed by recrystallization in aqueous acetic acid.[15] 4-Methyloxindole has been prepared by reported procedures.[16] 6-Fluorooxindole (**6**) was prepared starting from 2,5-difluoronitrobenzene according to literature procedures[17] with some modifications (Scheme 2). Displacement of *o*-fluoro substitution of 2,5-difluoronitrobenzene with dimethyl malonate followed by hydrolytic decarboxylation with 6 N aqueous hydrochloric acid gave 4-fluoro-2-nitrophenylacetic acid (**5**). Reductive cyclization of **5** under hydrogenation conditions resulted in the generation of **6**. 5-Bromooxindole (**7**) was prepared by bromination of oxindole with NBS in acetonitrile at 0 °C (Scheme 2). Synthesis of 5-methyloxindole (**8**) was achieved by a Wolff−Kishner-like reduction of 5-methylisatin with hydrazine hydrate (Scheme 2).[18] Most of the aldehydes (**9**−**12**) which are not commercially available were prepared by a Vilsmeier formylation reaction starting from commercially available chemicals (Scheme 3).

The 3-substituted indolin-2-ones may exist as either the *Z* or *E* isomer depending on the characteristics of the substituents at the C-3 position of the 3-substituted indolin-2-one (Tables 1−5). The two isomer forms could be distinguished by either 1D or 2D NOE analysis (Chart 2). The *Z*-configured compounds should show a NOE effect between the proton at the C-4 position and the vinyl proton, whereas the *E*-configured compounds should have a NOE effect between the proton at the C-4

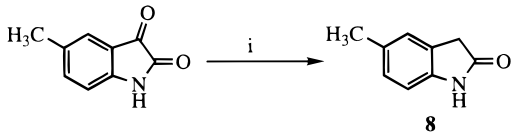
Scheme 2. Preparations of 6-Fluoro- (**6**), 5-Bromo- (**7**), and 5-Methyloxindoles (**8**)



i. $CH_2(COOCH_3)_2$/NaH/DMSO/100 °C/3 hr; ii. 6 N HClaq/reflux/2 days; iii. H_2/10% Pd-C/CH_3OH.

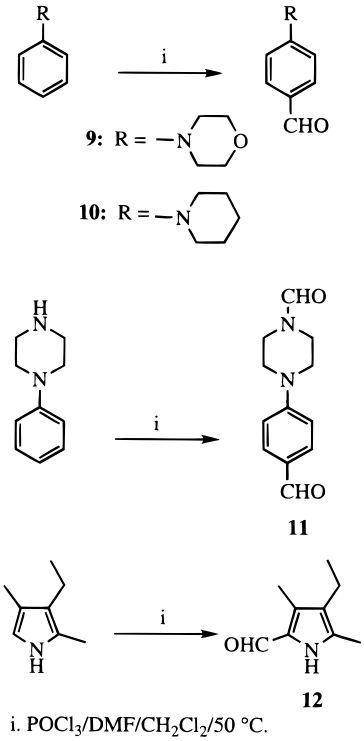


i. NBS/acetonitrile/0 °C/3 h.



i. $NH_2NH_2 \cdot H_2O$/100°C.

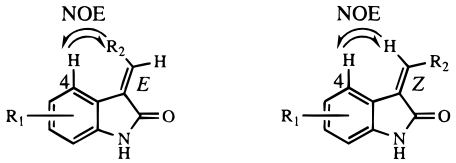
Scheme 3. Synthesis of the Corresponding Aldehydes



i. $POCl_3$/DMF/CH_2Cl_2/50 °C.

position and the proton(s) in the C-3 substitution of the 3-substituted indolin-2-ones. The configurations of some of the compounds (reference compounds) were determined using NOE analysis, while the configurations of the remaining compounds were assigned by comparison of their ^{1}H NMR spectrum with those of the reference compounds. The relationship between the *Z*−*E* configuration of a compound and the chemical

Chart 2. Determination of the Configurations for 3-Substituted Indolin-2-ones Using NOE Experiments

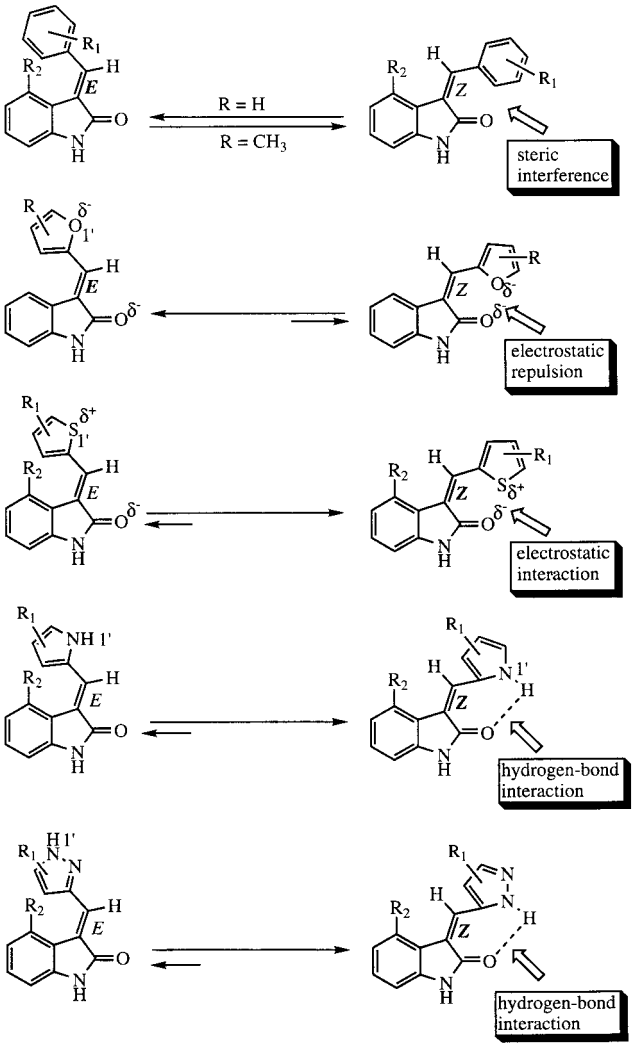


shifts of its particular proton(s) is summarized in Tables 1−5. For example, NOE experiments demonstrated that the chemical shifts for the protons at the C-2′ and C-6′ positions in the phenyl ring at the C-3 position of the 3-(substituted benzylidenyl)indolin-2-ones (Table 1) were around 7.85−8.53 ppm for the *Z* isomer but 7.45−7.84 ppm for the *E* isomer. We interpret this to be due to 2′- or 6′-protons being deshielded by the carbonyl at the C-2 position of the indolin-2-one ring in the *Z* isomer form. In the *E* isomer form, these protons may be shielded by the phenyl ring of the indolin-2-one core structure. Thus, configurations of the compounds within this series could be assigned based on the chemical shifts of particular protons, H-2′ and/or H-6′, in the molecules (Table 1).

Most of 3-(substituted benzylidenyl)indolin-2-ones which do not have any substitution at the C-4 position in general existed as the mixtures of both *Z* and *E* isomer forms. Compound **3** analogues (Table 2) containing a pyrrole substituent at the C-3 position of the 3-substituted indolin-2-ones existed exclusively as the *Z* isomer forms (Table 2). Indolin-2-ones having 3-substituted thienyl group (Table 3) existed as *Z* isomers. 3-[(Substituted pyrazolyl)methylidenyl]indolin-2-ones (Table 5) having a proton at the N-1′ position in the pyrazole ring (**62**) existed as the *Z* isomer, but the similar compound with substitution at the N-1′ position of the pyrazole ring existed as the *E* isomer (**63**). However, compounds with a 3-substituted furanyl moiety existed as *E* isomers (Table 4).

We have observed the equilibrium between the *Z* and *E* isomer forms in polar solvents, such as methanol and dimethyl sulfoxide (DMSO), or in the presence of light. The *E* isomerism of the 3-(substituted benzylidenyl)-indolin-2-ones in Table 1 may be the result of the steric interaction between the carbonyl group at the C-2 position of the indolin-2-one ring and the proton at the C-2′(6′) position of the phenyl ring in their *Z* isomeric forms (Scheme 4). 3-[(Substituted furanyl)methylidenyl]indolin-2-ones appear to favor the *E* isomer form. This may be due to the electrostatic repulsion between the C-2 carbonyl oxygen atom of the indolin-2-one and the O-1′ of furan in their *Z* isomer form (Scheme 4). On the other hand, the predominance of the *Z* isomerism of the 3-[(substituted pyrrolyl)methylidenyl]indolin-2-ones and 3-[(substituted pyrazolyl)methylidenyl]indolin-2-ones may be the result of the intramolecular hydrogen bonding between the C-2 carbonyl oxygen atom of the indolin-2-one ring and the proton on the N-1′ of pyrrole or N-2′ of pyrazole rings (Scheme 4). Replacement of the proton on the N-1′ position of the pyrrole (**50**) or pyrazole (**63**) ring afforded *E*-configured compounds due to the unfavorable interaction between the substitution at the N-1′ position of the pyrrole or the lone pair at the N-2′ position of the pyrazole ring and the carbonyl oxygen atom at the C-2 position of the indolin-2-one ring

Scheme 4. Rationales for the Preferred *Z* or *E* Configuration of 3-Substituted Indolin-2-ones



in the *Z*-configured form. The loss of the intramolecular hydrogen bonding of such compounds is also responsible for the *E* isomeric form. Electrostatic interaction between the C-2 carbonyl oxygen and partial positively charged S-1′ of the 3-[(substituted thienyl)methylidenyl]-indolin-2-ones favors the *Z* isomer form for the compounds in Table 3.

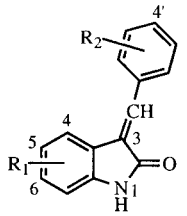
Results and Discussion

Structure−activity relationships (SAR) for inhibition of ligand-dependent tyrosine phosphorylation by the indolin-2-ones were determined in a panel of NIH3T3 mouse fibroblast lines engineered to overexpress various RTKs including the human PDGF receptor β, the murine VEGF receptor (*Flk-1*), the human EGF receptor, and the human insulin-like growth factor-1 (IGF-1) receptor.[19] To assess homology to the EGF receptor family-2 (Her-2) (p185^{erbB2}) kinase activity, the cytoplasmic domain of the human Her-2 was engineered to contain the extracellular and transmembrane domain of the human EGF receptor. The chimeric receptor was overexpressed in NIH3T3 cells, and ligand-dependent Her-2 kinase activity was assessed following stimulation of the receptor with EGF as described.[19] IC_{50} values were defined as the concentration of a compound required to achieve 50% inhibition of tyrosine phosphorylation on various RTKs compared to ligand-stimulated control reactions in the presence of vehicle alone (dimethyl sulfoxide). Compounds with IC_{50} values greater than 100 μM were considered inactive. The relative selectivity of a given inhibitor was assessed using calculated ratios of the IC_{50} values corresponding to specific RTKs. The results are summarized in Tables 1−5.

The primary goal of this study was to establish an initial SAR on these novel RTK inhibitors and to identify compounds with high potency and selectivity against particular RTKs as chemical leads. Five classes of compounds were designed and synthesized in order to provide answers to the following questions: (A) What are the electronic and steric effects of the substitutions in the phenyl ring of **1** or in the pyrrole ring of **3** at the C-3 position of 3-substituted indolin-2-ones on the inhibitory potency and specificity against various RTKs? (B) Do different heteroaryl substitutions at the C-3 position of 3-substituted indolin-2-ones influence the inhibitory activity of this series? (C) Is the *Z* isomer the active form? (D) Is the vinyl proton essential for the inhibitory activity? (E) What is the impact of the substituent(s) in the indolin-2-one core structure on potency and selectivity of the inhibitors against various RTKs.

A. SAR of 3-(Substituted benzylidenyl)indolin-2-ones. Compounds from Table 1 were synthesized and represented analogues of the two initial leads, **1** and **2**. These compounds possess substituents on the phenyl ring at the C-3 position of 3-substituted indolin-2-ones. As mentioned above, there were two differences when chemical structures of **1** and **2** were compared. The former has a nitrogen atom and the latter an isosteric methine attached to the C-4′ position of the phenyl group at the C-3 position of 3-substituted indolin-2-one. In addition, **1** exists as the *Z* isomer, but **2** exists predominantly as the *E* isomer. Compound **1** was shown to be a selective VEGF (*Flk-1*) RTK inhibitor, whereas **2** is a nonselective RTK inhibitor. These results suggested that electron-rich substitution (4′-dimethylamino in **1**) in the phenyl ring at the C-3 position of 3-substituted indolin-2-ones would have a large impact on the configuration and subsequently the inhibitory activity of inhibitors against various RTKs. On the basis of these preliminary results, we designed, synthesized, and evaluated a series of analogues of **1** with different electron-rich substituents at the C-4′ position of the phenyl ring at the C-3 position of 3-substituted indolin-2-ones. First we maintained the nitrogen atom at the C-4′ position of **1** and incorporated this nitrogen atom into the piperazine, morpholine, pyrrolidine, or piperidine substitution. Second, we replaced the 4′-dimethylamino substitution of **1** with other electron-rich substituents such as a hydroxyl or alkoxy moiety. Third, we introduced electron-withdrawing substituents into the phenyl ring at the C-3 position of the indolin-2-ones. Fourth, based on our knowledge of particular tyrphostins that have bulky substituents and had been shown to be EGF RTK inhibitors, bulky substituents, such as *tert*-butyl or isopropyl, were added on the phenyl at the C-3 position of the indolin-2-ones in order to achieve specific EGF

Table 1. 3-(Substituted benzylidenyl)indolin-2-ones: Preferred Configurations Determined by NMR Experiments and Their Inhibitory Activities toward Particular RTKs at the Cellular Level



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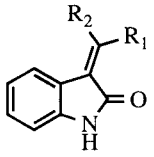
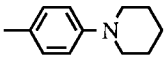


37

Compound	Substitutions		NMR Data chemical shift in ppm			Inhibition of Cellular Tyrosine Kinase Activity (IC_{50}, μM)				
ID	R_1	R_2	% of *Z*-isomer	*Z*-isomer 2',6'-H	*E*-isomer 2',6'-H	PDGF RTK	FLK-1 RTK	EGF RTK	HER-2 RTK	IGF-1 RTK
1	H	4'-$N(CH_3)_2$	100 (NOE)	8.44	N/A	19.4	0.8	>100	>100	>100
13	1-CH_3	4'-$N(CH_3)_2$	97	8.47	7.83	>100	22.6	>100	>100	>100
14	4-CH_3	4'-$N(CH_3)_2$	100 (NOE)	8.32	N/A	>100	3.0	>100	>100	>100
15	6-F	4'-$N(CH_3)_2$	20	8.39	7.61	66.0	4.6	>100	>100	>100
16	H	4'-N(piperazine)N-CHO	0 (NOE)	N/A	7.67	5.1	1.6	>100	90.2	>100
17	4-CH_3	4'-N(piperazine)N-CHO	97	8.26	7.73	24.5	2.5	>100	>100	67.6
18	5-Br	4'-N(piperazine)N-CHO	21	8.38	7.58	13.8	>50	>50	>50	>50
19	H	4'-N(morpholine)O	3	8.43	7.66	33.1	2.5	>100	>100	>100
20	4-CH_3	4'-N(morpholine)O	99 (NOE)	8.28	7.73	49.9	2.1	>100	66.6	>100
21	5-Cl	4'-N(morpholine)O	23	8.49	7.67	>10	4.1	>10	>10	>10
22	5-Br	4'-N(morpholine)O	23	8.38	7.57	>10	>10	>10	>10	>10
23	H	4'-N(pyrrolidine)	86	8.46	7.80	>100	3.0	>100	>100	>100
24	H	4'-N(piperidine)	4	8.43	7.64	>100	4.5	>100	92.6	>100
25	H	4'-OH	3	8.39	7.69	>100	2.7	>100	>100	>100
26	H	4'-OCH_3	4	8.45	7.69	>100	7.6	>100	>100	>100
27	H	4'-Br	3	8.30	7.70	>100	>100	>100	>100	>100
28	H	4'-COOH	0	N/A	7,80	>100	>100	>100	>100	>100
29	H	3', 5'-$C(CH_3)_3$ 4'-OH	32	8.39	7.53	>100	8.4	7.9	64.8	49.5
30	5-Cl	3', 5'-$CH(CH_3)_2$ 4'-OH	33	8.36	7.45	60.4	4.3	9.5	8.2	>100
31	H	3'- $C(CH_3)_3$ 4-OCH_3, 5'- Br	4	7.85 (6') 8.20(2')	7.68(6') 7.84(2')	>100	46.6	19.0	19.0	>100

Table 1 (Continued)

Compound	Substitutions		NMR Data chemical shift in ppm			Inhibition of Cellular Tyrosine Kinase Activity (IC_{50}, μM)				
ID	R_1	R_2	% of *Z*-isomer	*Z*-isomer 2',6'-H	*E*-isomer 2',6'-H	PDGF RTK	FLK-1 RTK	EGF RTK	HER-2 RTK	IGF-1 RTK
32	5-Cl	3'-$C(CH_3)_3$ 4'-OCH_3	93 (NOE)	8.53 (2') 8.41 (6')	7.62 (2') 7.57 (6')	>100	>100	19.3	16.2	>100
2	H	4'-$CH(CH_3)_2$	5 (NOE)	8.31	7.64	24.2	5.2	18.5	16.9	10.0
33	H	3', 5'-$CH(CH_3)_2$ 4'-OH	17	8.33	7.45	17.5	8.5	15.3	7.0	10.1
34	H	3'-$C(CH_3)_3$ 4'-OCH_3	9 (NOE)	8.51 (2') 8.38 (6')	7.56-7.64	>100	>100	>100	13.2	>100
35	1-CH_3	4'-Br	100	8.32	N/A	>100	>100	>100	22.5	>100
36	5-Cl	3'-$C(CH_3)_3$ 4'-OCH_3, 5'-Br	89 (NOE)	8.83 (2') 8.38 (6')	7.71 (2') 7.49 (6')	>100	>100	>100	15.2	>100
37	CH_3		0 (NOE)	--	--	>100	>100	>100	>100	>100

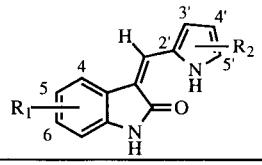
RTK inhibitors. Last, to assess the importance of the vinyl proton, we replaced the vinyl proton in compound **24** by a methyl group in compound **37**. The SAR analysis of these analogues of **1** is presented below.

We incorporated the 4′-nitrogen atom of **1** into different saturated heterocyclic rings to give **16** (4′-piperazine), **19** (4′-morpholine), **23** (4′-pyrrolidine), and **24** (4′-piperidine). Evaluation of these compounds indicated that they were inactive against the EGF, Her-2, and IGF-1 RTKs as is the case of **1**. Using the PDGF RTK assay, **16** was 4-fold more potent and **19** 2-fold less potent than **1**. In contrast, **23** and **24** were found to be inactive against the PDGF RTK activity. However, their inhibitory activity against the VEGF (*Flk-1*) RTK was 2−6-fold less than that of **1**. Interestingly, these compounds showed similar potency against the VEGF (*Flk-1*) RTK regardless of their predominant configurations. However, our cocrystal study indicated that the binding configuration for **16** was the *Z* form despite its predominantly *E* isomer in solution (Table 1).[14] Therefore, the *E* form of **16** has to be isomerized to the *Z* form before or during the interaction with the VEGF (*Flk-1*) RTK. Binding of the *Z* isomer to the catalytic site may shift the *Z*−*E* equilibrium from the *E* isomer to the *Z* form. This hypothesis may help to explain the loss of potencies of **16**, **19**, **23**, and **24** against the VEGF (*Flk-1*) RTK activity relative to **1** which exists only in the active *Z* form.

Since the *Z* configuration may be the preferred form for the inhibitory activity of these inhibitors against both the PDGF and VEGF (*Flk-1*) RTKs, we have designed and synthesized conformationally constrained 3-substituted indolin-2-ones. In this regard, a methyl group was introduced at the C-4 position of the 3-substituted indolin-2-ones in order to provide more steric interactions with the substituent at the C-3 position. In this case, the *Z* isomer form was more favored than the *E* form (Scheme 4). Compound **17** and **20** (Table 1) were prepared by introducing the methyl group at the C-4 position of **16** and **19** (Table 1), respectively. Both **17** and **20** existed exclusively as *Z* isomers compared to the predominant *E* isomer forms of their respective parent compounds, **16** and **19**. Compound **14**, while existing as a *Z* isomer like **1**, was found to be inactive against the PDGF RTK activity and about 4-fold less active (IC_{50} = 3.0 μM) than **1** (IC_{50} = 0.8 μM) against the VEGF (*Flk-1*) RTK activity. Therefore, substitution at the C-4 position of the indolin-2-one core is unfavorable for the inhibitory activities against both the PDGF and VEGF (*Flk-1*) RTKs. Compound **17** was found to be 5-fold less potent than **16** against the PDGF RTK activity. Compound **20** showed similar potency to **19** when tested against the PDGF and VEGF (*Flk-1*) RTK activities despite the unfavorable interaction introduced by the methyl substituent at the C-4 position. In conclusion, the actual inhibitory activities of these 3-substituted indolin-2-ones may depend on the balance between the positive inhibitory effect of the *Z*-configured piperazine ring of **17** or the piperidine ring of **20** and the negative inhibitory impact of the methyl group at the C-4 position.

Other electron-rich substituents, such as a hydroxyl or methoxy moiety (**25** and **26**, respectively) were also introduced at the C-4′ position of the phenyl substitution at the C-3 position of the 3-substituted indolin-2-ones. Compound **25** containing the hydroxyl group at the C-4′ position selectively inhibited the VEGF (*Flk-1*) RTK activity with an IC_{50} of 2.7 μM. Methylation of this hydroxyl substitution at the C-4′ position in **25** gave **26** which specifically inhibits VEGF (*Flk-1*) RTK activity with an IC_{50} value of 7.6 μM. In contrary, inhibitors having electron-withdrawing substituents at the C-4′ position of the phenyl ring at the C-3 position of 3-substituted indolin-2-ones totally eliminated the inhibitory activity against any RTK, as evidenced by **27** and **28**. This result suggests that an electron-rich substituent may be essential for the inhibitory activity against the VEGF (*Flk-1*) RTK, probably through an impact on the equilibrium between *Z* and *E* forms. Electron-rich substituents could establish a highly

Table 2. 3-[(Substituted pyrrol-2-yl)methylidenyl]indolin-2-ones: Preferred Configurations Determined by NMR Experiments and Their Inhibitory Activity toward Particular RTKs at the Cellular Level

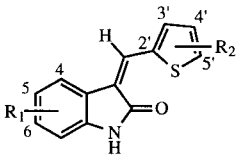


Compound	Substitutions		NMR Data chemical shift in ppm			Inhibition of Cellular Tyrosine Kinase Activity (IC_{50}, μM)				
ID	R_1	R_2	% of *Z*-isomer	H-vinyl	H-4	PDGF RTK	FLK-1 RTK	EGF RTK	HER-2 RTK	IGF-1 RTK
3	H	H	100 (NOE)	7.71	7.61	12.0	0.39	>100	>100	>100
38	4-CH_3	H	100 (NOE)	7.65	N/A	>100	31.0	>100	>100	>100
39	5-Cl	H	100 (NOE)	7.86	7.74	85.4	3.0	>100	>100	>100
40	6-F	H	100	7.69	7.61	2.8	<0.78	>100	>100	>100
41	H	3', 4'-CH_3	100 (NOE)	7.62	7.75	>100	2.7	>100	>100	>100
42	H	3'-CH_2CH_2COOH 4'-CH_3	100 (NOE)	7.70	7.68	60.9	0.40	>100	>100	>100
43	H	3'-CH_3 4'-$CH_2CH_2COOCH_3$	100 (NOE)	7.61	7.74	66.0	6.3	>100	>100	>100
44	H	3'-CH_3 4'-$COOCH_2CH_3$	100 (NOE) 45	7.72	N/A	>100	0.18	>100	>100	>100
45	H	3', 5'-CH_3	100 (NOE)	7.55	7.70	20.26	1.04	>100	>100	>100
46	6-F	3', 5'-CH_3	100	7.52	7.70	18.2	<0.78	>100	>100	>100
47	H	3', 5'-CH_3 4'-CH_2CH_3	100	7.54	7.69	>100	>100	>100	>100	>100
48	H	3', 5'-CH_3 4'-$COOCH_2CH_3$	100	7.62	7.80	>100	0.07	>100	>100	>100
49	H	3'-CH_2CH_3 4', 5'-CH_3	100 (NOE)	7.52	7.69	65.5	0.14	>100	>100	>100
50	H	1'-CH_3	0	7.47	7.98	>100	20.4	>100	>100	>100
51	5-NO_2	H	100 (NOE)	8.14	8.85	41.7	>100	>100	>100	>100
52	4-CH_3	3', 5'-CH_3	100 (NOE)	7.51	N/A	>100	1.0	>100	>100	>100
53	5-CH_3	3', 5'-CH_3	100	7.49	7.52	25.5	0.3	>100	>100	>100

conjugated system, decrease the energy barrier between the *Z* and *E* forms, and enable the *Z*−*E* equilibrium to be established faster than the one without such electron-rich substituents.

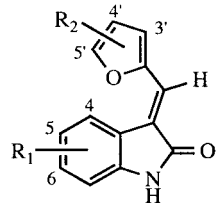
To examine the effects of the substituents around the indolin-2-one ring on RTK inhibitory potency and selectivity, **13**, **15**, **18**, **21**, and **22** have been synthesized. Methyl substitution at the N-1 position of **13** abolishes the inhibitory activity against the PDGF RTK activity and results in 28-fold loss in potency against the VEGF (*Flk-1*) RTK activity (IC_{50} = 22.6 μM) compared to **1**. This result indicated the importance of the proton at the N-1 position for inhibitory activity of **1** and is in agreement with our studies derived from the cocrystal structure of **16** and FGF RTK catalytic domain.[14] In this latter case, the proton at the N-1 position of **16** was

Table 3. 3-[(Substituted thien-2-yl)methylidenyl]indolin-2-ones: Preferred Configurations Determined by NMR Experiments and Their Inhibitory Activity toward Particular RTKs at the Cellular Level



Compound	Substitutions		NMR Data chemical shift in ppm			Inhibition of Cellular Tyrosine Kinase Activity (IC_{50}, μM)				
ID	R_1	R_2	% of Z-isomer	H-vinyl	H-4	PDGF RTK	FLK-1 RTK	EGF RTK	HER-2 RTK	IGF-1 RTK
54	H	3'-Br	100	7.49	7.52	>100	15.4	>100	>100	>100
55	H	4'-Br	100	8.04	7.59	>100	>100	>100	>100	>100
56	H	5'-SCH_3	100	7.98	7.64	>100	2.34	>100	>100	>100
57	4-CH_3	5'-SCH_3	100 (NOE)	7.87	N/A	>100	>100	>100	91.4	>100
58	H	5'-CH_2CH_3	100	7.98	7.64	>100	3.40	44.0	>100	>100

Table 4. 3-[(Substituted furan-2-yl)methylidenyl]indolin-2-ones: Preferred Configurations Determined by NMR Experiments and Their Inhibitory Activity toward Particular RTKs at the Cellular Level

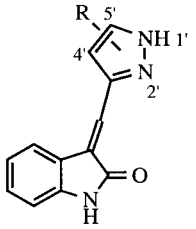


Compound	Substitutions		NMR Data chemical shift in ppm			Inhibition of Cellular Tyrosine Kinase Activity (IC_{50}, μM)				
ID	R_1	R_2	% of Z-isomer	H-vinyl	H-4	PDGF RTK	FLK-1 RTK	EGF RTK	HER-2 RTK	IGF-1 RTK
59	H	H	0 (NOE)	7.33	8.35	>100	>100	>100	>100	>100
60	H	5'-CH_3	0 (NOE)	7.24	8.29	>100	73.6	>100	>100	>100
61	H	5'-CH_2CH_3	0 (NOE)	7.25	8.31	>100	41.2	>100	>100	>100

found to be involved in a critical hydrogen-bonding interaction with the carbonyl oxygen of Glu562 within the catalytic domain.[14] Compound **15**, 6-fluoro-substituted **1**, was found to be 3.4- and 5.8-fold less potent than **1** against the PDGF and VEGF (*Flk-1*) RTK activities, respectively. Thus, the electron-withdrawing substituents on the indolin-2-one ring are unfavorable for inhibitory activity against both the PDGF and VEGF (*Flk-1*) RTKs.

To examine the effect of multiple substituents in the phenyl ring on the RTK inhibitory potency and selectivity, a series of analogues of **25** and **26** were prepared with substituents at the C-3′ and/or C-5′ position. Interestingly, the compound **25** analogue, **29** (which has *tert*-butyl at the C-3′ and C-5′ positions), was found to be active against not only the VEGF (*Flk-1*) RTK but also the EGF and Her-2 RTK activities compared to **25**. In addition, the compound **26** analogue, **34** (which has *tert*-butyl substitution at the C-3′ position), selectively inhibited the Her-2 RTK activity. Compound **33**, containing 3′,5′-diisopropyl moieties (compound **25** analogue), was found to be a nonselective RTK inhibitor. Of particular interest, **36**, the 5′-bromo analogue of the parent **32**, was a Her-2-specific inhibitor. Deleting the 5-chloro substitution of **32** resulted in **34** which was shown to specifically inhibit the Her-2 RTK activity. In contrary, deleting the 5-chloro substitution in compound **36** switched the selectivity from Her-2-specific to EGF/Her-2/VEGF (*Flk-1*) RTK-selective (**31**). The same 5-chloro substitution in different compounds has an

Table 5. 3-[(Substituted pyrazol-3-yl)methylidenyl]indolin-2-ones: Preferred Configurations Determined by NMR Experiments and Their Inhibitory Activity toward Particular RTKs at the Cellular Level



Compound		NMR Data chemical shift in ppm			Inhibition of Cellular Tyrosine Kinase Activity (IC_{50}, μM)				
ID	**R**	**% of Z-isomer**	**H-vinyl**	**H-4**	**PDGF RTK**	**FLK-1 RTK**	**EGF RTK**	**HER-2 RTK**	**IGF-1 RTK**
62	4'-Cl	100 (NOE)	7.53	7.87	>100	7.41	>100	>100	>100
63	1'-CH_3 4'-Cl	0 (NOE)	8.19	8.88	>100	>100	>100	>100	>100

opposite impact on the inhibitory activity of these inhibitors. This may be the result of different RTK conformational changes induced by inhibitors containing different structural features. A similar phenomenon was observed for the 4-(phenylamino)quinazolines.[9] In this series, specific inhibition of the EGF and Her-2 RTK family was accomplished with compounds that contain bulky lipophilic substitutions in the phenyl ring at the C-3 position of the 3-substituted indolin-2-ones, whereas selective inhibitors of the PDGF or/and VEGF (*Flk-1*) RTK family contained electron-rich substitutions in the phenyl ring.

To test the importance of the vinyl proton, compound **37**, an analogue of **24** (Table 1), was synthesized and evaluated. Both **37** and **24** exist as *E* isomers. However, compound **24**, a specific VEGF (*Flk-1*) RTK inhibitor, can be contrasted to **37** which was found to be inactive when tested against any RTK activity in the study. This demonstrates that the vinyl proton is critical for the inhibitory activity of these novel RTK inhibitors against VEGF (*Flk-1*) RTK.

B. 3-[(Substituted pyrrol-2-yl)methylidenyl]indolin-2-ones (Compound 3 Analogues). Since we have found that **3** inhibits both VEGF (*Flk-1*) (IC_{50} = 0.39 μM) and PDGF (IC_{50} = 12.0 μM) RTK activities, we designed a series of inhibitors with different five-membered rings at the C-3 position of 3-substituted indolin-2-ones, including pyrrole (Table 2), thiophene (Table 3), furan (Table 4), and pyrazole (Table 5). In the pyrrole series (Table 2), most of the compounds were found to exist as *Z* isomer forms and found to be inactive against the EGF, Her-2, and IGF-1 RTK activities. These compounds were found to be more potent and selective than 3-(substituted benzylidenyl)indolin-2-ones against the VEGF (*Flk-1*) RTK, and a few compounds showed high inhibitory activity against the PDGF RTK activity. Methylation at the N-1′ position of the pyrrole in **3** resulted in a compound (**50**) that existed as an *E* isomer and was shown to be a weak inhibitor of the VEGF (*Flk-1*) RTK. This suggested that the *Z* isomer is required for inhibiting the VEGF (*Flk-1*) RTK activity by these 3-substituted indolin-2-ones. This finding is in agreement with a cocrystal study of the indolin-2-ones and the FGF RTK catalytic domain.[14]

The roles of the substituents in the indolin-2-one core of these 3-[(substituted pyrrolyl)methylidenyl]indolin-2-ones in the inhibitory activity against the PDGF and VEGF (*Flk-1*) RTKs were evaluated. Compound **38**, the 4-methyl analogue of **3**, was found to be inactive when tested against the PDGF RTK activity (IC_{50} > 100 μM) and 79-fold less active (IC_{50} = 31.0 μM) than **3** when tested against the VEGF (*Flk-1*) RTK activity. Therefore, substitution at the C-4 position of the indolin-2-one ring was found to have a negative impact on the inhibitory activity of compounds when tested against both PDGF and VEGF (*Flk-1*) RTK activities. This is in accordance with conclusions following our SAR analysis of 3-(substituted benzylidenyl)indolin-2-ones as shown in Table 1. An electron-withdrawing chloro or fluoro substitution in the indolin-2-one core of **39** or **40** decreased the potency and selectivity when tested against the VEGF (*Flk-1*) RTK activity compared to **3**. The 5-nitro analogue (**51**) of **3** was found to be a weak PDGF RTK inhibitor and was found to be inactive when tested against the VEGF (*Flk-1*) RTK activity, suggesting that the electron-withdrawing substituents at the C-5 position of the indolin-2-ones may be detrimental to inhibitory activities associated with this series of RTK inhibitors.

To assess the impact of the substitutions on the pyrrole ring of **3** on the inhibitory activity against the VEGF (*Flk-1*) RTK, small alkyl or electron-withdrawing groups were introduced into the pyrrole ring. Inhibitory activities associated with compounds having small alkyl and electron-withdrawing polar moieties at both the C-3′ and C-4′ positions in the pyrrole ring of **3** were found to be either highly selective or specific toward the VEGF (*Flk-1*) RTK as in the case of **42** and **44**. On the other hand, most of the compounds with substitutions at the C-3′ and C-5′ positions in the pyrrole ring of **3** have shown decreased potency when tested against the PDGF RTK activity and maintained the good inhibitory activity when tested against the VEGF (*Flk-1*) RTK activity. Compound **45**, the 3′,5′-dimethyl analogue[20] of **3**, was

found to be slightly less potent than **3** when tested against the PDGF ($IC_{50} = 20.26\ \mu M$) and VEGF (*Flk-1*) RTK ($IC_{50} = 1.04\ \mu M$), respectively. Interestingly, the 4′-ethyl analogue (**47**) of **45** was found to be inactive when tested against all of the RTK activities in the panel, whereas the 4′-ethoxycarbonyl analogue (**48**) of **45** was found to be the most potent and selective VEGF (*Flk-1*) RTK inhibitor shown in Table 2. In contrast, compound **49** which has a 3′-ethyl-4′,5′-dimethylpyrrole at the C-3 position of the indolin-2-ones was found to be highly potent when tested against the VEGF (*Flk-1*) RTK activity ($IC_{50} = 0.14\ \mu M$) and weakly active when tested against the PDGF RTK activity. These results suggest that an electron-withdrawing polar substitution at the C-4′ position of the pyrrole ring of **3** is favorable for the VEGF (*Flk-1*) RTK inhibitory activity of these inhibitors. Therefore, adjustment of substituents at the C-4′ position could lead to highly potent and selective inhibitors of the VEGF (*Flk-1*) RTK. Compound **52**, the 4-methyl analogue of **45**, was found to be inactive when tested against the PDGF RTK activity and was found to be 3-fold more potent than **45** when tested against the VEGF (*Flk-1*) RTK activity. This result is in contrary to the one derived from the 3-(substituted benzylidenyl)indolin-2-ones. The methyl group at the C-4 position of **14** (Table 1) has negative impact on the potency toward both the PDGF and VEGF (*Flk-1*) RTKs. In addition, the methyl substitution at the C-4 position of **38** decreased the potency against both the PDGF and VEGF (*Flk-1*) RTKs when compared to **3**. Interestingly, a methyl substitution at the C-5 position of **53** was found to be 10-fold more potent than **45** when tested against the VEGF (*Flk-1*) RTK activities. In summary, these studies have shown that small electron-donating substituents at the C-4 or C-5 position were favorable for inhibitory activity of the compounds containing 3′,5′-dimethyl-substituted pyrrole at the C-3 position of the indolin-2-ones when tested against the VEGF (*Flk-1*) RTK.

C. 3-[(Substituted thien-2-yl)methylidenyl]indolin-2-ones. In the 3-[(substituted thien-2-yl)methylidenyl]indolin-2-one series (Table 3), we have determined that most of the compounds exist predominantly as *Z* isomers. However, these compounds, in general, are less potent than the 3-[(substituted pyrrol-2-yl)-methylidenyl]indolin-2-ones against the VEGF (*Flk-1*) RTK. Methyl substitution at the C-4 position of the indolin-2-one ring eliminated the VEGF (*Flk-1*) RTK inhibitory activity as in the case of **57** ($IC_{50} > 100\ \mu M$) when compared to the parent compound **56** ($IC_{50} = 2.30\ \mu M$).

D. 3-[(Substituted furan-2-yl)methylidenyl]indolin-2-ones. 3-[(Substituted furan-2-yl)methylidenyl]-indolin-2-ones (Table 4) were found to exist as *E* isomers. Given our model, it is not surprising that these compounds were found to be weak inhibitors of the VEGF (*Flk-1*) RTK and inactive against other RTK activities measured. In addition, the furan ring is less electron-rich than a pyrrole or thiophene ring. Therefore, these electronic effects may also influence the inhibitory activity of these compounds.

E. 3-[(Substituted pyrazol-3-yl)methylidenyl]-indolin-2-ones. In the series of 3-[(substituted pyrazol-3-yl)methylidenyl]indolin-2-ones (Table 5), compound **62** was shown to specifically inhibit the VEGF (*Flk-1*) RTK activity ($IC_{50} = 7.41\ \mu M$). Alkylation at the N-1′ position in the pyrazole ring of **62** resulted in **63** which was found to be inactive against any RTK activity measured. NOE NMR experiments indicated that **62** exists as the *Z* isomer whereas **63** exists as the *E* isomer. We suggest that the proton at the N-1′ position of the pyrazole ring in **62** may migrate to the N-2′ position and allow intramolecular hydrogen bonding with the carbonyl oxygen atom at the C-2 position of the indolin-2-one (Scheme 4). Such hydrogen bonding may be responsible for the *Z* configuration of **62**. Methylation at the N-1′ position of the pyrazole ring in **62** may disrupt the hydrogen bonding and result in a repulsive interaction between the lone pair electrons at the N-2′ position of the pyrazole ring and the carbonyl oxygen at the C-2 position of the indolin-2-one. This model may help to explain why **63** exists as the *E* isomer and why these two compounds may have differences in inhibitory activity against the VEGF (*Flk-1*) RTK.

Conclusions

This report describes the identification, characterization, and SAR analysis of a novel chemical series of tyrosine kinase inhibitors, 3-substituted indolin-2-ones, which exhibit selectivity toward particular RTKs. In this report, the potency and selectivity of an inhibitor using cell-based assays can be further influenced by many factors including vehicle solubility, membrane permeability, chemical stability and metabolism, and alternative targets that may affect RTK activation. Nonetheless, SAR analysis can be very useful using these assays to identify chemical leads when performed with compounds that have similar physical and chemical properties. In this regard, we have shown that the potency and selectivity of this chemical series depend to a large extent on the configuration of the compound as it relates to substitutions at the C-3 position of the indolin-2-one core. The 3-[(substituted pyrrolyl-, thienyl-, and pyrazolyl)methylidenyl]indolin-2-ones were shown to exist exclusively or predominantly as *Z* isomers and were also found to be highly selective toward the VEGF (*Flk-1*) and/or PDGF RTK activity while inactive or weakly active against the EGF, Her-2, and IGF-1 RTK activities. The association of these inhibitory activities with the *Z* isomeric forms was found to be in agreement with our cocrystal studies using **16** and **42** bound in the catalytic core of the FGF RTK.[14] In this latter study, the binding configurations for both **16** and **42** were found to be the *Z* isomer forms. Since the primary amino acid sequences of the VEGF (*Flk-1*) and PDGF receptor kinase domains are highly homologous to that of the FGF receptor kinase, we suggest that the ability of many of the indolin-2-ones to show specific inhibitory properties on the VEGF (*Flk-1*) and PDGF RTKs may be due to the existence of *Z* isomers that can bind into the ATP binding pocket of these RTKs. In addition, the same study showed that the pyrrole ring and the indolin-2-one ring in **42** are near coplanar due to the intramolecular hydrogen bonding between the proton at the N-1′ position and oxygen atom at the C-2 position of **42**. The cocrystal X-ray structures of the catalytic domain of the FGF receptor with **16** and **42** suggest that the 3-substituted indolin-2-one compounds may bind at the ATP binding site with the indolin-2-

one ring occupying the adenine binding pocket. Bidentate hydrogen bonding between the indolin-2-one flat core structure in the adenine binding site may play a crucial role to block entry of ATP in the binding site. Both the proton at the N-1 position and the oxygen atom at the C-2 position of the indolin-2-one were found to be coordinated to the peptide backbone within the adenine binding cleft. In this regard, alkylation at the N-1 position of the indolin-2-ones greatly decreases the inhibitory potency of these 3-substituted indolin-2-ones (**13** vs **1**) against both the PDGF and VEGF (*Flk-1*) RTKs.

In addition, we have suggested that the substitution around the indolin-2-ones may be the key determinants for the potency and especially specificity of the inhibitors. A fluoro substitution at the C-6 position of the 3-[(substituted pyrrolyl)methylidenyl]indolin-2-ones greatly increased the inhibitory activity against the PDGF RTK (**40** vs **3**). A chloro substitution at the C-5 position of the 3-[(substituted pyrrolyl)methylidenyl]-indolin-2-one decreased inhibitory activity against both the PDGF and VEGF (*Flk-1*) RTKs (**39** vs **3**). A strong electron-withdrawing nitro group was detrimental to the inhibitory activity of **51** against the VEGF (*Flk-1*) RTK. It was also observed that same substitution could have different impacts on the inhibitory activity of different inhibitors depending on other substitutions in the molecules. In this regard, fluoro substitution at the C-6 position of **15** decreased the inhibitory activity against both the PDGF and VEGF (*Flk-1*) RTK activities when compared to **1**. This may imply different conformational changes of the enzyme induced by different inhibitors. Therefore, different SAR conclusions have been drawn for different types of 3-substituted indolin-2-ones. On the other hand, some of the 3-(substituted benzylidenyl)-indolin-2-ones that were found to be selective for both the EGF and Her-2 receptors possessed bulky lipophilic substitutions in the phenyl ring at the C-3 position of 3-(substituted benzylidenyl)indolin-2-ones. This result may indicate structural differences in the ATP binding sites when catalytic domains of the EGF and Her-2 RTKs are compared to those of the PDGF and VEGF (*Flk-1*) RTKs.

In summary of the SAR studies: (1) Proton at the N-1 position of the indolin-2-ones is essential for the inhibitory activity against the PDGF and VEGF (*Flk-1*) RTKs. (2) Vinyl proton is critical for 3-substituted indolin-2-ones to inhibit the PDGF and VEGF (*Flk-1*) RTKs. (3) *Z* Isomeric form is required for the inhibitory activity against the PDGF and VEGF (*Flk-1*) RTKs. (4) 3-[(Substituted pyrrolyl)methylidenyl]indolin-2-ones are the most potent and selective inhibitors of the PDGF and VEGF (*Flk-1*) RTKs. (5) Electron-donating substitution is preferred for the inhibitory potency of these 3-substituted indolin-2-ones against the PDGF and VEGF (*Flk-1*) RTKs. (6) Bulky lipophilic groups in the phenyl ring at the C-3 position of 3-(substituted benzylidenyl)indolin-2-ones are the key determinants for the selective inhibitory activity of these inhibitors against the EGF and Her-2 RTKs.

In conclusion, we described a chemical series that shows promise in the development of RTK inhibitors with the capacity to be specific to particular enzymes. PTK function has been associated with a wide variety of human diseases including cancers, metastasis, arterial restenosis, various inflammatory diseases including psoriasis and rheumatoid arthritis, pulmonary fibrosis, liver cirrhosis, kidney sclerosis, and other diseases. The potential to design and synthesize 3-substituted indolin-2-ones with inhibitory properties for various PTK subtypes supports the use of compounds of this class to treat a wide variety of human diseases.

Experimental Section

NMR spectra were recorded by Acorn NMR using a Nicolet NT300 or a Nicolet NT360 spectrometer. Tetramethylsilane (TMS) was used as an internal standard and chemical shifts are reported in parts per million (δ) downfield from TMS. Coupling constants are reported in hertz. Mass spectra (electron spray) were recorded by SYNPEP CORP, using an API I PLUS spectrometer. Elemental analyses were performed by Galbraith Laboratories, Inc. Elemental analyses results are within ±0.4% of the theoretical values.

General Procedure for 3-(Substituted benzylidenyl)-indolin-2-one Analogues (Compound 1 Analogues). A reaction mixture of the proper oxindole (1 equiv), aldehyde (1.2 equiv), and piperidine (0.1 equiv) in ethanol 1−2 mL/1 μmol oxindole) was stirred at 90 °C for 3−5 h. After the mixture cooled, the precipitate was filtered, washed with cold ethanol, and dried to give the target compound in over 80% yield. The analytical data reported here is for the major isomer. The data for the minor isomer is not reported. The ratios of *Z* to *E* isomers for each compound are shown in Table 1.

(*Z*)-3-[4-(Dimethylamino)benzylidenyl]indolin-2-one (1): ^{1}H NMR (300 MHz, DMSO-d_6) δ 10.46 (s, 1H, NH-1), 8.44 (d, J = 9.24 Hz, 2H, H-2′,6′), 7.60 (s, 1H, H-vinyl), 7.59 (d, J = 7.82 Hz, 1H, H-4), 7.11 (dt, J = 1.02, 7.82 Hz, 1H, H-6), 6.93 (dt, J = 0.89, 7.82 Hz, 1H, H-5), 6.80 (d, J = 7.82 Hz, 1H, H-7), 6.73 (d, J = 9.24 Hz, 2H, H-3′,5′), 3.00 (s, 6H, N(CH$_3$)$_3$-4′); MS *m*/*z* (relative intensity, %) 256 (100, [M + 1]$^{+\bullet}$). Anal. ($C_{17}H_{16}N_2O\cdot 0.1H_2O$) C, H, N.

(*Z*)-1-Methyl-3-[4-(dimethylamino)benzylidenyl]indolin-2-one (13): ^{1}H NMR (360 MHz, DMSO-d_6) δ 8.47 (d, J = 9.15 Hz, 2H, H-2′,6′), 7.67 (s, 1H, H-vinyl), 7.66 (d, J = 7.79 Hz, 1H, H-4), 7.21 (dt, J = 1.09, 7.79 Hz, 1H, H-6), 7.02 (dt, J = 0.99, 7.79 Hz, 1H, H-5), 6.95 (d, J = 7.79 Hz, 1H, H-7), 6.78 (d, J = 9.15 Hz, 2H, H-3′,5′), 3.22 (s, 3H, NCH$_3$-1), 3.05 (s, 6H, N(CH$_3$)$_2$-4′); MS *m*/*z* (relative intensity, %) 279 (100, [M + 1]$^{+\bullet}$), 349 (100, [M + 1]$^{+\bullet}$). Anal. ($C_{18}H_{18}N_2O$) C, H, N.

(*Z*)-4-Methyl-3-[4-(dimethylamino)benzylidenyl]indolin-2-one (14): ^{1}H NMR (360 MHz, DMSO-d_6) δ 10.40 (s, br, 1H, NH-1), 8.32 (d, J = 8.24 Hz, 2H, H-2′,6′), 7.61 (s, 1H, H-vinyl), 7.02 (t, J = 7.81 Hz, 1H, H-6), 6.67−6.75 (m, 4H, H-5,7,3′,5′), 3.02 (s, 6H, N(CH$_3$)$_2$-4′); MS *m*/*z* (relative intensity, %) 278 (100, M$^{+\bullet}$). Anal. ($C_{18}H_{18}N_2O\cdot 0.1H_2O$) C, H, N.

(*E*)-6-Fluoro-3-[4-(dimethylamino)benzylidenyl]indolin-2-one (15). 2-(4-Fluoro-2-nitrophenyl)malonic Acid Dimethyl Ester (4). The reaction mixture of sodium hydride (2.6 g, 108 μmol) and 14.5 g of dimethyl malonate (108 μmol) in 160 mL of dimethyl sulfoxide was heated to 100 °C for 1 h. The mixture was cooled to room temperature, and 7.95 g of 2,5-difluoronitrobenzene (70 μmol) was added and stirred for 30 min. The mixture was then heated to 100 °C for 1 h, cooled to room temperature, and poured into 400 mL of saturated ammonium chloride solution. The mixture was extracted with 200 mL of ethyl acetate and the organic layer washed with brine, dried over anhydrous sodium sulfate, and concentrated under vacuum. The residue was crystallized from methanol to give 24.4 g (80% yield) of dimethyl 4-fluoro-2-nitrophenylmalonate as a white solid. The filtrate was concentrated and chromatographed on a column of silica gel (ethyl acetate/hexane, 1:8) to give 5.03 g of dimethyl 4-fluoro-2-nitrophenylmalonate (96% total yield): ^{1}H NMR (360 MHz, DMSO-d_6) δ 8.02 (dd, J = 8.54, 2.48 Hz, 1H, H-3), 7.68 (dt, J = 8.54, 2.48 Hz, 1H, H-5), 7.63 (dd, J = 8.54, 5.63 Hz, 1H, H-6), 5.47 (s, 1H, C*H*(COOCH$_3$)$_2$-1), 3.70 (s, 6H, CH(COOC*H*$_3$)$_2$-1).

Dimethyl 4-fluoro-2-nitrophenylmalonate (5.0 g, 18.5 μmol) was refluxed in 20 mL of 6 N hydrochloric acid for 24 h. The reaction mixture was cooled and the white solid collected by vacuum filtration, washed with water, and dried to give 3.3 g (87% yield) of 4-fluoro-2-nitrophenylacetic acid (**5**): ^{1}H NMR (360 MHz, DMSO-d_6) δ 12.5 (s, br, 1H, CH_2COOH-1), 7.96 (d, J = 10.21 Hz, 1H, H-3), 7.58−7.61 (m, 2H, H-5,6), and 3.97 (s, 2H, CH_2COOH-1).

4-Fluoro-2-nitrophenylacetatic acid (3.3 g, 16.6 μmol) dissolved in 15 mL of acetic acid was hydrogenated over 0.45 g of 10% palladium on carbon under 60 psi for 2 h. The catalyst was removed by filtration and washed with 15 mL of methanol. The combined filtrates were concentrated and diluted with water. The precipitate was collected by vacuum filtration, washed with water, and dried to give 1.6 g (70% yield) of 6-fluoroindolin-2-one (**6**): ^{1}H NMR (360 MHz, DMSO-d_6) δ 10.43 (s, br, 1H, NH-1), 7.17 (t, J = 8.13 Hz, 1H, H-4), 6.69 (ddd, J = 2.22, 8.13, 10.38 Hz, 1H, H-5), 6.60 (dd, J = 2.22, 9.16, Hz, 1H, H-7), 3.42 (s, 2H, CH_2-3); MS *m*/*z* (relative intensity, %) 153 (100, $[M + 1]^{+\bullet}$).

Preparation of 15. The title compound was prepared by condensation of **6** and (dimethylamino)benzaldehyde using the same method described for analogues of **1**: ^{1}H NMR (360 MHz, DMSO-d_6) δ 10.56 (s, 1H, NH-1), 7.76 (dd, J = 7.69, 8.41 Hz, 1H, H-4), 7.61 (d, J = 8.79 Hz, 2H, H-2′,6′), 7.49 (s, 1H, H-vinyl), 6.80 (d, J = 8.79 Hz, 2H, H-3′,5′), 6.78−6.64 (m, 2H, H-5,7), 3.01 (s, 6H, $N(CH_3)_2$-4′); MS *m*/*z* (relative intensity, %) 283 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{17}H_{15}FN_2O$) C, H, N.

Preparation of (*E*)-3-[4-(1-Formylpiperazin-4-yl)benzylidenyl]indolin-2-one (16). 4-(1-Formylpiperazin-4-yl)-benzaldehyde (11). To a solution of 30 mL (0.3 mol) of dimethylformamide in 20 mL of anhydrous 1,2-dichloroethane was added dropwise 30 mL (0.3 mol) of phosphorus oxychloride at 0 °C. The ice bath was removed. The reaction mixture was stirred for an additional 30 min and cooled in an ice bath. 1-Phenylpiperazine (16.0 g, 0.1 μmol) was added to the above solution portionwise over 15 min, and the reaction mixture was stirred at 50 °C for 1 h. The reaction mixture was poured into ice-cold 1 N sodium hydroxide solution and stirred at room temperature for 1 h. The organic layer was separated, and the aqueous layer was extracted with ethyl acetate. The combined organic layer was washed with brine until pH = 7, dried over anhydrous sodium sulfate, and evaporated. The residue was separated on a silica gel column eluting with a mixture of ethyl acetate and hexane to afford 9.0 g (41%) of the title compound as a light-yellow solid: MS (ES) *m*/*z* (relative intensity, %) 217 ($[M - 1]^{+\bullet}$, 100); ^{1}H NMR (360 MHz, DMSO) δ 9.74 (s, 1H), 8.11 (s, 1H), 7.73 (d, J = 9.1 Hz, 2H), 7.08 (d, J = 9.1 Hz, 2H), 3.39−3.53 (m, 8H).

(*E*)-3-[4-(1-Formylpiperazin-4-yl)benzylidenyl]indolin-2-one (16). A reaction mixture of 133.15 mg (1 μmol) of oxindole, 228.3 mg (1.2 μmol) of **11**, and 3 drops of piperidine in 2 mL of ethanol was stirred at 90 °C for 5 h. After the mixture cooled, the precipitate was filtered, washed with cold ethanol, and dried to yield 199.5 mg (65%) of the title compound as a yellow solid: MS (ES) *m*/*z* (relative intensity, %) 333 ($M^{+\bullet}$, 17); ^{1}H NMR (360 MHz, DMSO) δ 10.52 (s, 1H, NH-1), 8.11 (s, 1H, CHO-4″), 7.75 (d, J = 7.3 Hz, 1H, H-4), 7.67 (d, J = 8.7 Hz, 2H, H-2′,6′), 7.55 (s, 1H, H-vinyl), 7.21 (t, J = 7.3 Hz, 1H, H-5), 7.10 (d, J = 8.7 Hz, 2H, H-3′,5′), 6.87−6.92 (m, 2H, H-6,7), 3.31−3.56 (m, 8H). Anal. ($C_{20}H_{19}N_3O_2$) C, H, N.

(*Z*)-3-[4-(1-Formylpiperazin-4-yl)benzylidenyl]-4-methylindolin-2-one (17): ^{1}H NMR (360 MHz, DMSO) δ 10.42 (s, 1H, NH-1), 8.26 (d, J = 9.14 Hz, 2H, H-2′,6′), 8.09 (s, 1H, H-vinyl), 7.60 (s, 1H, CHO-1′), 7.03 (t, J = 7.75 Hz, 1H, H-6), 7.00 (d, J = 9.14 Hz, 2H, H-3′,5′), 6.74 (d, J = 7.75 Hz, 1H, H-5), 6.66 (d, J = 7.75 Hz, 1H, H-7), 3.51 (m, 4-H, H-2′,6′), 3.36 (m, 4H, H-3′,5′); MS (ES) *m*/*z* (relative intensity, %) 348 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{21}H_{21}N_3O_2 \cdot 0.2H_2O$) C, H, N.

(*E*)-5-Bromo-3-[4-(1-formylpiperazin-4-yl)benzylidenyl]-indolin-2-one (18). Preparation of 5-Bromoindolin-2-one (7). Oxindole (1.3 g, 10 μmol) in 20 mL of acetonitrile was cooled to −10 °C, and 2.0 g of *N*-bromosuccinimide (10 μmol) was slowly added with stirring. The reaction mixture was stirred for 1 h at −10 °C and for 2 h at 0 °C. The precipitate was collected, washed with water, and dried to give 1.9 g (90%yield) of 5-bromoindolin-2-one.

Synthesis of 18. The title compound was synthesized from **7** and 4-(1-formylpiperazin-4-yl)benzaldehyde by the same condensation method described for analogues of **1**: ^{1}H NMR (360 MHz, DMSO) δ 10.54 (s, 1H, NH-1), 8.04 (s, 1H, H-vinyl), 7.73 (d, J = 1.87 Hz, 1H, H-4), 7.58 (d, J = 8.76 Hz, 2H, H-2′,6′), 7.53 (s, 1H, CHO-4″), 7.30 (dd, J = 1.87, 8.27 Hz, 1H, H-6), 7.05 (d, J = 8.74 Hz, 2H, H-3′5′), 6.77 (d, J = 8.27 Hz, 1H, H-7), 3.44−3.48 (m, 2H, H-2″,6″), 3.27−3.57 (m, 2H, H-3″,5″). Anal. ($C_{20}H_{18}BrN_3O_2$) C, H, N.

Preparation of (*E*)-3-[4-(Morpholin-4-yl)benzylidenyl]-indolin-2-one (19). 4-(Morpholin-4-yl)benzaldehyde (9). To a solution of 15 mL of dimethylformamide in 50 mL of 1,2-dichloroethane was added dropwise 10 mL of phosphorus oxychloride at 0 °C. The ice bath was removed, and the reaction mixture was stirred for an additional 30 min. 4-Phenylmorpholine (16.3 g) was added to the above solution portionwise, and the reaction mixture was refluxed for 2 days. Triethylamine (2.5 mL) was added to the above reaction mixture, and the reaction mixture was refluxed for 2 days. The reaction mixture was poured into ice-cold 1 N sodium hydroxide solution (pH = 9 after mixing), and the resulting mixture was stirred at room temperature for 1 h. The organic layer was separated, and the aqueous layer was extracted with 2 × 20 mL of dichloromethane. The combined organic layer was washed with brine until pH = 7, dried over anhydrous sodium sulfate, and evaporated. The residue was separated on a silica gel column eluting with a solvent mixture of ethyl acetate and hexane to afford 12.95 g (68%) of the title compound as a white solid.

(*E*)-3-[4-(Morpholin-4-yl)benzylidenyl]indolin-2-one (19). A reaction mixture of 6.66 g of oxindole, 11.50 g of **9**, and 5 mL of piperidine in 50 mL of ethanol was stirred at 90 °C for 5 h. After cooling, the precipitate was filtered, washed with cold ethanol, and dried to yield 15.0 g (98%) of the title compound as a yellow solid: ^{1}H NMR (360 MHz, DMSO-d_6) δ 10.45 (s, 1H, NH-1), 7.74 (d, J = 7.20 Hz, 1H, H-4), 7.66 (d, J = 8.98 Hz, 2H, H-2′,6′), 7.54 (s, 1H, H-vinyl), 7.20 (t, J = 7.20 Hz, 1H, H-6), 7.05 (d, J = 8.98 Hz, 2H, H-3′,5′), 6.86−6.90 (m, 2H, H-5,7), 3.75 (t, J = 4.77 Hz, 4H, H-2″,4″), 3.28 (t, J = 4.77 Hz, 4H, H-3″,5″); MS *m*/*z* (relative intensity, %) 307 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{19}H_{18}N_2O_2$) C, H, N.

(*Z*)-4-Methyl-3-[4-(morpholin-4-yl)benzylidenyl]indolin-2-one (20): ^{1}H NMR (360 MHz, DMSO-d_6) δ 10.44 (s, 1H, NH-1), 8.28 (d, J = 8.98 Hz, 2H, H-2′,6′), 7.62 (s, 1H, H-vinyl), 7.04 (t, J = 7.69 Hz, 1H, H-6), 6.96 (d, J = 8.98 Hz, 2H, H-3′,5′), 6.76 (d, J = 7.69 Hz, 1H, H-5), 6.68 (d, J = 7.69 Hz, 1H, H-7), 3.74 (t, J = 4.98 Hz, 4H, H-2″,6″), 3.28 (t, J = 4.98 Hz, 4H, H-3″,5″), 2.57 (s, 1H, CH_3-4); 2D-NOE ^{1}H NMR showed the correlation between, H-vinyl and CH_3-4, which confirmed the *Z* conformer; MS *m*/*z* (relative intensity, %) 320 (100, $M^{+\bullet}$). Anal. ($C_{20}H_{20}N_2O_2$) C, H, N.

(*E*)-5-Chloro-3-[4-(morpholin-4-yl)benzylidenyl]indolin-2-one (21): ^{1}H NMR (360 MHz, DMSO-d_6) δ 110.59 (s, 1H, NH-1), 7.67 (d, J = 2.33 Hz, 1H, H-4), 7.66 (s, 1H, H-vinyl), 7.62 (d, J = 9.91 Hz, 2H, H-2′,6′), 7.25 (dd, J = 2.33, 8.20 Hz, 1H, H-6), 7.09 (d, J = 9.91 Hz, 2H, H-3′,5′), 6.88 (d, J = 8.20 Hz, 1H, H-7), 3.74−3.77 (m, 4H, H-2″,4″), 3.30−3.33 (m, 4H, H-3″,5″). Anal. ($C_{19}H_{17}ClN_2O_2 \cdot 0.6H_2O$) C, H, N.

(*E*)-5-Bromo-3-[4-(morpholin-4-yl)benzylidenyl]indolin-2-one (22): ^{1}H NMR (360 MHz, DMSO-d_6) δ 10.53 (s, br, 1H, NH-1), 7.73 (d, J = 1.96 Hz, 1H, H-4), 7.57 (d, J = 9.25 Hz, 2H, H-2′,6′), 7.53 (s, 1H, H-vinyl), 7.30 (dd, J = 1.96, 8.33 Hz, 1H, H-6), 7.01 (d, J = 9.25 Hz, 2H, H-3′,5′), 6.76 (d, J = 8.33 Hz, 1H, H-7), 3.67 (m, 4H, H-2″,6″), 3.25 (m, 4H, H-3″,5″); MS *m*/*z* (relative intensity, %) 386 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{19}H_{17}BrN_2O_2$) C, H, N.

(*Z*)-3-[4-(Pyrrolidin-1-yl)benzylidenyl]indolin-2-one (23): ^{1}H NMR (300 MHz, DMSO-d_6) δ 10.46 (s, 1H, NH-1), 8.46 (d, J = 9.14 Hz, 2H, H-2′,6′), 7.62 (s, 1H, H-vinyl), 7.61 (d, J =

8.21 Hz, 1H, H-4), 7.10 (dt, J = 1.09, 8.21 Hz, 1H, H-6), 6.93 (dt, J = 1.02, 7.65 Hz, 1H, H-5), 6.79 (d, J = 7.65 Hz, 1H, H-7), 6.61 (d, J = 9.14 Hz, 2H, H-3′,5′), 3.33−3.37 (m, 2H, H-2″,5″), 1.95−2.00 (m, 2H, H-3″,4″). Anal. ($C_{19}H_{18}N_2O$) C, H, N.

Synthesis of (*E*)-3-[4-(Piperidin-1-yl)benzylidenyl]indolin-2-one (24). 4-(Piperidin-1-yl)benzaldehyde (10). To a solution of 2.3 mL of dimethylformamide in 10 mL of 1,2-dichloroethane was added dropwise 2.8 mL of phosphorus oxychloride at 0 °C. The ice bath was removed, and the reaction mixture was stirred for 15 min. 1-Phenylpiperidine (3.2 mL) was added to the above solution portionwise, and the reaction mixture was refluxed overnight. The reaction mixture was poured into ice-cold 2 N sodium hydroxide solution and stirred at room temperature for 1 h. The organic layer was separated, and the aqueous layer was extracted with 2 × 20 mL of ethyl acetate. The combined organic layer was washed with brine until pH = 7, dried over anhydrous sodium sulfate, and evaporated. The residue was separated on a silica gel column eluting with ethyl acetate and hexane to afford 1.5 g (40%) of the title compound as a white solid.

(*E*)-3-[4-(Piperidin-1-yl)benzylidenyl]indolin-2-one (24). A reaction mixture of 134.0 mg of oxindole, 226.8 g of **10**, and 3 drops of piperidine in 2 mL of ethanol was stirred at 90 °C for 5 h. After the mixture cooled, the precipitate was filtered, washed with cold ethanol, and dried to yield 268.5 mg (88%) of the title compound as a yellow solid: ^{1}H NMR (300 MHz, DMSO-d_6) δ 10.48 9 (s, 1H, NH-1), 7.77 (d, J = 7.69 Hz, 1H, H-4), 7.64 (d, J = 8.91 Hz, 2H, H-2′,6′), 7.51 (s, 1H, H-vinyl), 7.19 (dt, J = 1.12, 7.69 Hz, 1H, H-6), 7.03 (d, J = 8.91 Hz, 2H, H-3′,5′), 6.89 (dt, J = 0.78, 7.69 Hz, 1H, H-5), 6.86 (d, J = 7.69 Hz, 1H, H-7), 3.36 (s, br, 4H, CH_2-2″,6″), 1.60 (s, br, 6H, CH_2-3″,4″,5″); MS m/z (relative intensity, %) 305 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{20}H_{20}N_2O$) C, H, N.

(*E*)-3-(4-Hydroxybenzylidenyl)indolin-2-one (25): ^{1}H NMR (360 MHz, DMSO-d_6) δ 10.46 (s, 1H, NH-1), 10.06 (s, br, 1H, OH-4′), 7.69 (d, J = 7.52 Hz, 1H, H-4), 7.62 (d, J = 8.54 Hz, 2H, H-2′,6′), 7.54 (s, 1H, H-vinyl), 7.21 (dt, J = 1.09, 7.52 Hz, 1H, H-6), 6.86−6.93 (m, 4H, H-3′,5′,5,7); MS m/z (relative intensity, %) 238 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{15}H_{11}NO_2$) C, H, N.

(*E*)-3-(4-Methoxybenzylidenyl)indolin-2-one (26): ^{1}H NMR (360 MHz, DMSO-d_6) δ 10.48 (s, 1H, NH-1), 7.69 (d, J = 8.82 Hz, 2H, H-2′,6′), 7.64 (d, J = 7.55 Hz, 1H, H-4), 7.57 (s, 1H, H-vinyl), 7.20 (t, J = 7.55 Hz, 1H, H-6), 7.08 (d, J = 8.82 Hz, 2H, H-3′,5′), 6.83−6.88 (m, 2H, H-5,7), 3.83 (s, 3H, OCH_3-4′); MS m/z (relative intensity, %) 252 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{16}H_{13}NO_2$) C, H, N.

(*E*)-3-(4-Bromobenzylidenyl)indolin-2-one (27): ^{1}H NMR (300 MHz, DMSO-d_6) δ 10.61 (s, 1H, NH-1), 7.70 (d, J = 8.63 Hz, 2H, H-2′,6′), 7.63 (d, J = 8.63 Hz, 2H, H-3′,5′), 7.55 (s, 1H, H-vinyl), 7.47 (d, J = 7.58 Hz, 1H, H-4), 7.22 (t, J = 7.58 Hz, 1H, H-6), 6.81−6.87 (m, 2H, H-5,7); MS m/z (relative intensity, %) 300 (100, $M^{+\bullet}$). Anal. ($C_{15}H_{10}BrNO\cdot 0.3H_2O$) C, H, N.

(*E*)-3-(4-Carboxybenzylidenyl)indolin-2-one (28): ^{1}H NMR (360 MHz, DMSO-d_6) δ 13.08 (s, br, 1H, COOH-4′), 10.61 (s, br, 1H, NH-1), 8.07 (d, J = 8.06 Hz, 2H), 7.80 (d, J = 8.06 Hz, 2H), 7.65 (s, 1H, H-vinyl), 7.45 (d, J = 7.51 Hz, 1H, H-4), 7.25 (dt, J = 0.94, 7.53 Hz, 1H, H-6), 6.83−6.90 (m, 2H, H-5,7). Anal. ($C_{16}H_{11}NO_3\cdot 0.125H_2O$) C, H, N.

(*E*)-3-(3,5-Di-*tert*-butyl-4-hydroxybenzylidenyl)indolin-2-one (29): ^{1}H NMR (360 MHz, DMSO-d_6) δ 10.43 (s, 1H, NH-1), 7.58 (s, 1H, H-vinyl), 7.57 (d, J = 6.80 Hz, 1H, H-4), 7.53 (s, 2H, H-2′,6′), 7.19 (t, J = 6.80 Hz, 1H, H-6), 6.85−6.89 (m, 2H, H-5,7), 1.41 (s, 18H, $C(CH_3)_3$-3′,5′); MS m/z (relative intensity, %) 350 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{23}H_{27}NO_2$) C, H, N.

(*E*)-3-(3,5-Diisopropyl-4-hydroxybenzylidenyl)-5-chloroindolin-2-one (30): ^{1}H NMR (300 MHz, DMSO-d_6) δ 10.64 (s, 1H, NH-1), 8.94 (s, br, 1H, OH-4′), 7.73 (d, J = 1.96 Hz, 1H, H-4), 7.66 (s, 1H, H-vinyl), 7.45 (s, 2H, H-2′,6′), 7.26 (dd, J = 1.96, 8.36 Hz, 1H, H-6), 6.89 (d, J = 8.36 Hz, 1H, H-7), 3.31−3.42 (m, 2H, C*H*$(CH_3)_2$-3′,5′), 1.21 (d, J = 6.51 Hz, 12H, $CH(CH_3)_2$-3′,5′); MS m/z (relative intensity, %) 356 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{21}H_{22}ClNO_2$) C, H, N.

(*E*)-3-(3-Bromo-5-*tert*-butyl-4-methoxybenzylidenyl)indolin-2-one (31): ^{1}H NMR (360 MHz, DMSO-d_6) δ 10.64 (s, 1H, NH-1), 7.84 (d, J = 2.02 Hz, H-2′), 7.68 (d, J = 2.02 Hz, 1H, H-6′), 7.54 (s, 1H, H-vinyl), 7.51 (d, J = 7.69 Hz, 1H, H-4), 7.26 (dt, J = 1.29, 7.75 Hz, 1H, H-6), 6.85−6.87 (m, 2H, H-5,7), 3.93 (s, 3H, OCH_3-4′), 1.37 (s, 9H, $C(CH_3)_3$-5′); MS m/z (relative intensity, %) 388 (100, $[M + 2]^{+\bullet}$). Anal. ($C_{20}H_{20}BrNO_2\cdot 0.5H_2O$) C, H, N.

(*Z*)-5-Chloro-3-(3-*tert*-butyl-4-methoxybenzylidenyl)indolin-2-one (32): ^{1}H NMR (360 MHz, DMSO-d_6) δ 10.58 (s, 1H, NH-1), 8.53 (d, J = 2.13 Hz, 1H, H-2′), 8.41 (dd, J = 2.13, 8.44 Hz, 1H, H-6′), 7.89 (s, 1H, H-vinyl), 7.81 (d, J = 2.22 Hz, 1H, H-4), 7.18 (dd, J = 2.22, 8.26 Hz, 1H, H-6), 7.10 (d, J = 8.44 Hz, 1H, H-5′), 6.81 (d, J = 8.26 Hz, 1H, H-7), 3.90 (s, 3H, OCH_3-4′), 1.35 (s, 9H, $C(CH_3)_3$-3′); MS m/z (relative intensity, %) 342 (100, $M^{+\bullet}$). Anal. ($C_{20}H_{20}ClNO_2$) C, H, N.

(*E*)-3-(4-Isopropylbenzylidenyl)indolin-2-one (2): ^{1}H NMR (300 MHz, DMSO-d_6) δ 10.57 (s, 1H, NH-1), 7.59−7.65 (m, 4H, H-4,vinyl,2′,6′), 7.38 (d, J = 8.49 Hz, 2H, H-3′,5′), 7.21 (dt, J = 0.91, 7.82 Hz, 1H, H-6), 6.83−6.88 (m, 2H, H-5,7), 2.94 (h, J = 6.99 Hz, 1H, C*H*$(CH_3)_2$-4′), 1.23 (d, J = 6.99 Hz, 6H, CH(C*H*$_3)_2$-4′); MS m/z (relative intensity, %) 264 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{18}H_{17}NO$) C, H, N.

(*E*)-3-(3,5-Diisopropyl-4-hydroxybenzylidenyl)indolin-2-one (33): ^{1}H NMR (300 MHz, DMSO-d_6) δ 10.54 (s, 1H, NH-1), 8.87 (s, br, 1H, OH-4′), 7.74 (d, J = 7.47 Hz, 1H, H-4), 7.58 (s, 1H, H-vinyl), 7.45 (s, 2H, H-2′,6′), 7.20 (dt, J = 1.10, 8.37 Hz, 1H, H-6), 6.88 (d, J = 7.47 Hz, 1H, H-5), 6.87 (dt, J = 1.03, 8.37 Hz, 1H, H-7), 3.37 (m, 2H, C*H*$(CH_3)_2$-3′,5′), 1.20 (d, J = 6.77 Hz, 12H, CH(C*H*$_3)_2$-3′,5′). Anal. ($C_{21}H_{23}NO_2$) C, H, N.

(*E*)-3-(3-*tert*-Butyl-4-methoxybenzylidenyl)indolin-2-one (34): ^{1}H NMR (360 MHz, DMSO-d_6) δ 10.50 (s, 1H, NH-1), 7.66 (d, J = 7.82 Hz, 1H, H-4), 7.58−7.62 (m, 3H, H-vinyl and H-2′,6′), 7.20 (t, J = 7.65 Hz, 1H, H-6), 7.13 (d, J = 8.26 Hz, 1H, H-5′), 6.84−6.88 (m, 2H, H-5,7), 3.89 (s, 3H, OCH_3-4′), 1.35 (s, 9H, $C(CH_3)_3$-3′); MS m/z (relative intensity, %) 308 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{20}H_{21}NO_2$) C, H, N.

(*Z*)-3-(4-Bromobenzylidenyl)-1-methylindolin-2-one (35): ^{1}H NMR (300 MHz, DMSO-d_6) δ 8.33 (d, J = 8.56 Hz, 2H, H-2′,6′), 7.83 (s, 1H, H-vinyl), 7.75 (d, J = 7.42 Hz, 1H, H-4), 7.67 (d, J = 8.56 Hz, 2H, H-3′,5′), 7.33 (t, J = 7.42 Hz, 1H, H-6), 7.07 (t, J = 7.70 Hz, 1H, H-5), 7.00 (d, J = 7.70 Hz, 1H, H-7), 3.20 (s, 3H, NCH_3-1); MS m/z (relative intensity, %) 315 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{16}H_{12}BrNO$) C, H, N.

(*E*)-3-(3-Bromo-5-*tert*-butyl-4-methoxybenzylidenyl)-5-chloroindolin-2-one (36): ^{1}H NMR (360 MHz, DMSO-d_6) δ 10.70 (s, 1H, NH-1), 8.83 (d, J = 2.15 Hz, 1H, H-2′), 8.38 (d, J = 2.15 Hz, 1H, H-6′), 7.90 (s, 1H, H-vinyl), 7.82 (d, J = 1.97 Hz, 1H, H-4), 7.30 (dd, J = 1.97, 8.16 Hz, 1H, H-6), 6.83 (d, J = 8.16 Hz, 1H, H-7), 3.92 (s, 3H, OCH_3-4′), 1.39 (s, 9H, $C(CH_3)_3$-3′); MS m/z (relative intensity, %) 420 (100, $M^{+\bullet}$). Anal. ($C_{20}H_{19}BrClNO_2$) C, H, N.

(*E*)-3-[1-[4-(Piperidin-1-yl)phenyl]ethylidenyl]indolin-2-one (37). The reaction mixture of 142.3 mg of pyrrolidine (2.0 μmol) and 243.9 mg of 4-(piperidin-1-yl)phenylacetone (1.2 μmol) was refluxed for 1 h in toluene. A Dean−Stark trap filled with anhydrous sodium sulfate was used to remove water. Oxindole (133.0 mg, 1.0 μmol) was added to the above mixture, and the reaction mixture was stirred for an additional 5 h. Toluene was removed by a rotary pump, and the residue was separated on a silica gel column eluting with ethyl acetate and hexane to give 63.0 mg of the title compound (20%); ^{1}H NMR (360 MHz, DMSO-d_6) δ 10.43 (s, 1H, NH-1), 7.21 (d, J = 8.70 Hz, 2H, H-2′,6′), 7.04 (dt, J = 1.27, 7.72 Hz, 1H, H-6), 7.01 (d, J = 8.70 Hz, 2H, H-3′,5′), 6.76 (d, J = 7.72 Hz, 1H, H-7), 6.59 (dt, J = 1.12, 7.72 Hz, 1H, H-5), 6.44 (d, J = 7.72 Hz, 1H, H-4), 3.25 (t, J = 5.39 Hz, 4H, CH_2-2″,6″), 2.66 (s, 3H, CH_3-vinyl), 1.59−1.64 (m, 6H, CH_2-3″,4″,5″). MS m/z (relative intensity, %) 319 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{21}H_{22}N_2O\cdot 0.6H_2O$) C, H, N.

General Procedure for 3-[(Substituted pyrrolyl)methylidenyl]indolin-2-one Analogues (Compound 3 Analogues). A reaction mixture of the proper oxindole (indolin-2-one) (1 equiv), the appropriate aldehyde (1.2 equiv), and piperidine (0.1 μmol) in ethanol (1−2 mL/1 μmol oxindole) was stirred at 90 °C for 3−5 h. After the mixture cooled, the precipitate was filtered, washed with cold ethanol, and dried to give the target compound in over 80% yield. The analytical data reported here belongs to the major isomer. The data for the minor isomer is not reported. The ratios of *Z* to *E* isomers for each compound are shown in Tables 2−6.

(*Z*)-3-[(Pyrrol-2-yl)methylidenyl]indolin-2-one (3): ^{1}H NMR (360 MHz, DMSO-d_6) δ 13.34 (s, br, 1H, NH-1′), 10.83 (s, 1H, NH-1), 7.71 (s, 1H, H-vinyl), 7.61 (d, J = 7.68 Hz, 1H, H-4), 7.33 (s, br, 1H, H-5′), 7.14 (t, J = 7.68 Hz, 1H, H-6), 6.98 (t, J = 7.68 Hz, 1H, H-5), 6.88 (d, J = 7.68 Hz, 1H, H-7), 6.83 (t, J = 1.71 Hz, 1H, H-3′), 6.33−6.36 (m, 1H, H-4′); MS *m*/*z* (relative intensity, %) 211 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{13}H_{10}N_2O$) C, H, N.

(*Z*)-4-Methyl-3-[(pyrrol-2-yl)methylidenyl]indolin-2-one (38): ^{1}H NMR (360 MHz, DMSO-d_6) δ 13.50 (s, br, 1H, NH-1′), 10.87 (s, 1H, NH-1), 7.65 (s, 1H, H-vinyl), 7.32 (d, J = 1.68 Hz, 1H, H-3′), 7.05 (t, J = 7.76 Hz, 1H, H-6), 6.91 (t, J = 1.63 Hz, 1H, H-5′), 6.80 (d, J = 7.66 Hz, 1H, H-7), 6.76 (t, J = 7.76 Hz, 1H, H-5), 6.33−6.36 (m, 1H, H-4′), 2.57 (s, 3H, CH_3-4); MS *m*/*z* (relative intensity, %) 247 (100, $[M + Na]^{+\bullet}$). Anal. ($C_{14}H_{12}N_2O_3 \cdot 0.1H_2O$) C, H, N.

(*Z*)-5-Chloro-3-[(pyrrol-2-yl)methylidenyl]indolin-2-one (39): ^{1}H NMR (360 MHz, DMSO-d_6) δ 13.30 (s, br, 1H, NH-1′), 10.93 (s, 1H, NH-1), 7.86 (s, 1H, H-vinyl), 7.74 (d, J = 2.27 Hz, 1H, H-4), 7.38−7.40 (m, 1H, H-5′), 7.16 (dd, J = 2.27, 8.28 Hz, 1H, H-6), 6.88 (d, J = 8.28 Hz, 1H, H-7), 6.85 (dd, J = 1.59, 3.44 Hz, 1H, H-3′), 6.37−6.39 (m, 1H, H-4′); MS *m*/*z* (relative intensity, %) 245 (100, $M^{+\bullet}$). Anal. ($C_{13}H_9N_2OCl$) C, H, N.

(*Z*)-6-Fluoro-3-[(pyrrol-2-yl)methylidenyl]indolin-2-one (40): ^{1}H NMR (360 MHz, DMSO-d_6) δ 13.19 (s, 1H, NH-1′), 10.98 (s, 1H, NH-1), 7.69 (s, 1H, H-vinyl), 7.61 (dd, J = 5.47, 8.34 Hz, 1H, H-4), 7.32−7.33 (m, 1H, H-5′), 6.76−6.82 (m, 2H, H-5,3′), 7.69 (dd, J = 2.31, 9.19 Hz, 1H, H-7), 6.33−6.35 (m, 1H, H-4′); MS *m*/*z* (relative intensity, %) 229 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{13}H_9FN_2O$) C, H, N.

(*Z*)-3-[(3,4-Dimethylpyrrol-2-yl)methylidenyl]indolin-2-one (41). A reaction mixture of 67.0 mg of oxindole, 73.0 mg of the 3,4-dimethylpyrrole-2-carboxaldehyde, and 2 drops of piperidine in 2 mL of ethanol was stirred at 90 °C for 3 h. After the mixture cooled, the precipitate was filtered, washed with cold ethanol, and dried to yield 87.7 mg (37%) of the title compound as a yellow solid: ^{1}H NMR (300 MHz, DMSO-d_6) δ 13.28 (s, br, 1H, NH-1′), 10.82 (s, 1H, NH-1), 7.75 (d, J = 7.62 Hz, 1H, H-4), 7.62 (s, 1H, H-vinyl), 7.09−7.14 (m, 2H, H-6,5′), 6.98 (dt, J = 1.03, 7.62 Hz, 1H, H-5), 6.87 (d, J = 7.62 Hz, 1H, H-7), 2.25 (s, 3H, CH_3-4′), 2.02 (s, 3H, CH_3-3′); MS *m*/*z* (relative intensity, %) 239 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{15}H_{14}N_2O$) C, H, N.

(*Z*)-3-[(3-(2-Carboxyethyl)-4-methylpyrrol-2-yl)methylidenyl]indolin-2-one (42). A reaction mixture of 134.0 mg (1.0 μmol) of oxindole, 217.43 mg (1.2 μmol) of the 3-(2-carboxyethyl)-4-methylpyrrole-2-carboxaldehyde, and 3 drops of piperidine in 3 mL of ethanol was stirred at 90 °C for 3 h. After the mixture cooled, the precipitate was filtered, washed with cold ethanol, and dried to yield 172.4 mg (58%) of the title compound as a yellow solid: ^{1}H NMR (300 MHz, DMSO-d_6) δ 13.27 (s, 1H, CH_2CH_2COO*H*-3′), 10.84 (s, br, 1H, NH-1), 7.67−7.70 (m, 2H, H-4, vinyl), 7.08−7.13 (m, 2H, H-6,5′), 6.97 (dt, J = 0.99, 7.47 Hz, 1H, H-5), 6.86 (d, J = 7.47 Hz, 1H, H-7), 2.92 (t, J = 7.37 Hz, 2H, CH_2C*H*$_2$COOH-3′), 2.32 (t, J = 7.37 Hz, 2H, C*H*$_2CH_2$COOH-3′), 2.03 (s, 3H, CH_3-4′); MS *m*/*z* (relative intensity, %) 297 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{17}H_{16}N_2O_3$) C, H, N.

(*Z*)-3-[[3-(2-(Methoxycarbonyl)ethyl)-4-methylpyrrol-5-yl]methylidenyl]indolin-2-one (43): ^{1}H NMR (360 MHz, DMSO-d_6) δ 13.31 (s, br, 1H, NH-1′), 10.78 (s, br, 1H, NH-1), 7.74 (d, J = 7.59 Hz, 1H, H-4), 7.61 (s, 1H, H-vinyl), 7.09−7.14 (m, 2H, H-6,5′), 6.98 (dt, J = 0.84, 7.59 Hz, 1H, H-5), 6.87 (d, J = 7.59 Hz, 1H, H-7), 3.60 (s, 3H, CH_2CH_2COOC*H*$_3$-3′), 2.69 (t, J = 7.29 Hz, 2H, CH_2C*H*$_2$COOCH$_3$-4′), 2.56 (t, J = 7.29 Hz, 2H, C*H*$_2CH_2$COOCH$_3$-3′), 2.26 (s, 3H, CH_3-4′); MS *m*/*z* (relative intensity, %) 311 (56, $[M + 1]^{+\bullet}$). Anal. ($C_{18}H_{18}N_2O_3$) C, H, N.

(*Z*)-3-[[3-(Ethoxycarbonyl)-4-methylpyrrol-5-yl]methylidenyl]indolin-2-one (44): ^{1}H NMR (300 MHz, DMSO-d_6) δ 13.81 (s, br, 1H, NH-1′), 11.04 (s, br, 1H, NH-1), 7.84−7.85 (m, 2H, H-4,2′), 7.72 (s, 1H, H-vinyl), 7.18 (dt, J = 1.34, 7.70 Hz, 1H, H-6), 7.02 (dt, J = 1.02, 7.70 Hz, 1H, H-5), 6.90 (d, J = 7.70 Hz, 1H, H-7), 4.22 (q, J = 7.05 Hz, $COOC$*H*$_2CH_3$-3′), 2.54 (s, 3H, CH_3-4′), 1.29 (t, J = 7.05 Hz, 3H, $COOCH_2C$*H*$_3$-3′); MS *m*/*z* (relative intensity, %) 297 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{17}H_{16}N_2O_3$) C, H, N.

(*Z*)-3-[(2,4-Dimethylpyrrol-5-yl)methylidenyl]indolin-2-one (45). A reaction mixture of 134.0 mg of oxindole, 147.8 mg of the 3,5-dimethylpyrrole-2-carboxaldehyde, and 3 drops of piperidine in 2 mL of ethanol was stirred at 90 °C for 3 h. After the mixture cooled, the precipitate was filtered, washed with cold ethanol, and dried to yield 136.7 mg (57%) of the title compound as a yellow solid: ^{1}H NMR (300 MHz, DMSO-d_6) δ 13.36 (s, br, 1H, NH-1′), 10.77 (s, 1H, NH-1), 7.71 (d, J = 7.70 Hz, 1H, H-4), 7.55 (s, 1H, H-vinyl), 7.09 (dt, J = 1.24, 7.70 Hz, 1H, H-6), 6.97 (dt, J = 0.97, 7.70 Hz, 1H, H-5), 6.87 (d, J = 7.66 Hz, 1H, H-7), 6.00 (d, J = 2.25 Hz, 1H, H-3′), 2.32 (s, 3H, CH_3-4′(2′)), 2.30 (s, 3H, CH_3-2′(4′)); MS *m*/*z* (relative intensity, %) 239 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{15}H_{14}N_2O$) C, H, N.

(*Z*)-6-Fluoro-3-[(2,4-dimethylpyrrol-5-yl)methylidenyl]-indolin-2-one (46): ^{1}H NMR (300 MHz, DMSO-d_6) δ 13.19 (s, br, 1H, NH-1′), 10.85 (s, 1H, NH-1), 7.00 (dd, J = 5.48, 8.38 Hz, 1H, H-4), 7.52 (s, 1H, H-vinyl), 6.74−6.79 (m, 1H, H-5), 6.67 (dd, J = 2.43, 9.17 Hz, 1H, H-7), 5.98 (d, J = 2.41 Hz, 1H, H-3′), 2.30 (s, 3H, CH_3-2′), 2.23 (s, 3H, CH_3-4′); MS *m*/*z* (relative intensity, %) 257 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{15}H_{13}FN_2O$) C, H, N.

(*Z*)-3-[(3-Ethyl-2,4-dimethylpyrrol-5-yl)methylidenyl]-indolin-2-one (47). Preparation of 3,5-Dimethyl-4-ethylpyrrole-2-carboxaldehyde (13). To a solution of 2.3 mL (30 mol) of dimethylformamide in 50 mL of anhydrous 1,2-dichloroethane was added dropwise 2.8 mL (30 mol) of phosphorus oxychloride at 0 °C. The ice bath was removed. The reaction mixture was further stirred for 30 min, and cooled with ice bath. 2,5-Dimethyl-4-ethylpyrrole (2.46 g, 20 μmol) was added to the above solution portionwise, and the reaction mixture was stirred at 50 °C for 3 h. The reaction mixture was poured into ice-cold 1 N sodium hydroxide solution and stirred at room temperature for 2 h. The gray precipitate was filtered and washed with water to a pH of 7. The solid was dried in an oven overnight to give **13** (2.42 g, 80%): ^{1}H NMR (360 MHz, DMSO) δ 11.33 (s, br, 1H, CHO-2), 9.42 (s, 1H, NH-1), 2.38 (q, J = 7.55 Hz, 2H, C*H*$_2CH_3$-4), 2.20 (s, 3H, CH_3-3), 2.15 (s, 3H, CH_3-5), 0.99 (t, J = 7.55 Hz, 3H, CH_2C*H*$_3$-4).

Preparation of 47. Condensation of **13** with oxindole using the same procedure for analogues of **3** gave **47**: ^{1}H NMR (360 MHz, DMSO-d_6) δ 13.38 (s, br, 1H, NH-1′), 10.69 (s, 1H, NH-1), 7.69 (d, J = 7.34 Hz, 1H, H-4), 7.54 (s, 1H, H-vinyl), 7.07 (dt, J = 1.12, 7.34 Hz, 1H, H-6), 6.96 (dt, J = 1.07, 7.34 Hz, 1H, H-5), 6.86 (d, J = 7.34 Hz, 1H, H-7), 2.40 (q, J = 7.61 Hz, 2H, C*H*$_2CH_3$-3′), 2.29 (s, 3H, CH_3-2′(4′)), 2.25 (s, 3H, CH_3-4′-(2′)), 1.04 (t, J = 7.61 Hz, 3H, CH_2C*H*$_3$-3′); MS *m*/*z* (relative intensity, %) 267 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{17}H_{18}N_2O$) C, H, N.

(*Z*)-3-[(2,4-Dimethyl-3-(ethoxycarbonyl)pyrrol-5-yl)-methylidenyl]indolin-2-one (48). A reaction mixture of 134.0 mg of oxindole, 234.3 mg of 4-(ethoxycarbonyl)-3,5-dimethylpyrrole-2-carboxaldehyde, and 3 drops of piperidine in 3 mL of ethanol was stirred at 90 °C for 3 h. After the mixture cooled, the precipitate was filtered, washed with cold ethanol, and dried to yield 244.6 mg (79%) of the title compound as a yellow solid: ^{1}H NMR (300 MHz, DMSO-d_6) δ 13.88 (s, br, 1H, NH-1′), 10.95 (s, br, 1H, NH-1), 7.80 (d, J = 7.53 Hz, 1H, H-4), 7.62 (s, 1H, H-vinyl), 7.15 (dt, J = 1.10,

7.53 Hz, 2H, H-6), 7.00 (dt, J = 1.06, 7.53 Hz, 1H, H-5), 6.89 (d, J = 7.53 Hz, 1H, H-7), 4.21 (q, J = 7.32 Hz, 2H, COOC*H*2CH3-3′), 2.54 (s, 3H, CH_3-4′(2′)), 2.49 (s, 3H, CH_3-2′(4′)), 1.30 (t, J = 7.32 Hz, 3H, $COOCH_2CH_3$-3′); MS m/z (relative intensity, %) 311 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{18}H_{18}N_2O_3$) C, H, N.

(*Z*)-3-[(4-Ethyl-2,3-dimethylpyrrol-5-yl)methylidenyl]-indolin-2-one (49): ^{1}H NMR (360 MHz, DMSO-d_6) δ 13.39 (s, br, 1H, NH-1′), 10.69 (s, 1H, NH-1), 7.69 (d, J = 7.67 Hz, 1H, H-4), 7.52 (s, 1H, H-vinyl), 7.07 (dt, J = 1.11, 7.67 Hz, 1H, H-6), 6.96 (dt, J = 1.17, 7.67 Hz, 1H, H-5), 6.86 (d, J = 7.67 Hz, 1H, H-7), 2.70 (q, J = 7.47 Hz, 2H, CH_2CH_3-4′), 2.27 (s, 3H, CH_3-2′(3′)), 1.97 (s, 3H, CH_3-3′(2′)), 1.11 (t, J = 7.47 Hz, 3H, CH_2CH_3-4′); MS m/z (relative intensity, %) 267 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{17}H_{18}N_2O$) C, H, N.

(*Z*)-3-[(1-Methylpyrrol-5-yl)methylidenyl]indolin-2-one (50): ^{1}H NMR (360 MHz, DMSO-d_6) δ 10.45 (s, 1H, NH-1), 7.98 (d, J = 7.38 Hz, 1H, H-4), 7.47 (s, 1H, H-vinyl), 7.17−7.22 (m, 2H, H-6,5′), 7.05 (dd, J = 0.88, 3.82 Hz, 1H, H-3′), 6.95 (dt, J = 1.08, 7.38 Hz, 1H, H-5), 6.88 (d, J = 7.38 Hz, 1H, H-7), 6.31 (dd, J = 2.73, 3.82 Hz, 1H, H-4′), 3.78 (s, 3H, NCH_3-1′); MS m/z (relative intensity, %) 225 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{14}H_{12}N_2O$) C, H, N.

(*Z*)-5-Nitro-3-[(pyrrol-2-yl)methylidenyl]indolin-2-one (51): ^{1}H NMR (360 MHz, DMSO-d_6) δ 13.16 (s, br, 1H, NH-1′), 11.49 (s, 1H, NH-1), 8.58 (d, J = 2.10 Hz, 1H, H-4), 8.14 (s, 1H, H-vinyl), 8.08 (dd, J = 2.10, 8.51 Hz, 1H, H-6), 7.47 (dd, J = 2.67, 3.66 Hz, 1H, H-5′), 7.05 (d, J = 8.51 Hz, 1H, H-7), 6.97−6.98 (m, 1H, H-3′), 6.42−6.44 (m, 1H, H-4′); MS m/z (relative intensity, %) 255 (100, $M^{+\bullet}$). Anal. ($C_{13}H_9N_3O_3 \cdot 0.1H_2O$) C, H, N.

(*Z*)-4-Methyl-3-[(2,4-dimethylpyrrol-5-yl)methylidenyl]-indolin-2-one (52): ^{1}H NMR (360 MHz, DMSO-d_6) δ 10.75 (s, br, NH-1), 7.51 (s, 1H, H-vinyl), 7.00 (t, J = 7.65 Hz, 1H, H-6), 6.79 (d, J = 7.93 Hz, 1H, H-5), 6.75 (d, J = 7.93 Hz, 1H, H-7), 6.01 (d, J = 2.81 Hz, 1H, H-3′), 2.57 (s, 3H, CH_3-4), 2.32 (s, 3H, CH_3-2′), 2.24 (s, 3H, CH_3-4′); MS m/z (relative intensity, %) 253 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{16}H_{16}N_2O \cdot 0.3H_2O$) C, H, N.

(*Z*)-5-Methyl-3-[(2,4-dimethylpyrrol-5-yl)methylidenyl]-indolin-2-one (53). Preparation of 5-Methylindolin-2-one (8). 5-Methylisatin (15.0 g, 10.2 μmol) and 60 mL of hydrazine hydrate were heated to 140−160 °C for 4 h. The reaction mixture was cooled to room temperature, poured into 300 mL of ice water, and acidified to pH 2 with 6 N hydrochloric acid. After standing at room temperature for 2 days the precipitate was collected by vacuum filtration, washed with water, and dried under vacuum to give 6.5 g (47% yield) of 5-methylindolin-2-one: ^{1}H NMR (360 MHz, DMSO-d_6) δ 10.19 (s, 1H, NH-1), 6.98 (s, 1H, H-4), 6.94 (d, J = 8.11 Hz, 1H, H-6), 6.68 (d, J = 8.11 Hz, 1H, H-7), 3.39 (s, 2H, CH_2-3), and 2.22 (s, 3H, CH_3-5).

Synthesis of 53. The title compound was prepared in the same way as **3** as described above: ^{1}H NMR (300 MHz, DMSO-d_6) δ 13.35 (s, br, 1H, NH-1′), 10.64 (s, 1H, NH-1), 7.52 (s, 1H, H-4), 7.49 (s, 1H, H-vinyl), 6.89 (d, J = 7.72 Hz, 1H, H-6), 6.73 (d, J = 7.72 Hz, 1H, H-7), 5.98 (d, J = 1.91 Hz, 1H, H-3′), 2.29 (s, 9H, CH_3-5,2′,4); MS m/z (relative intensity, %) 253 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{16}H_{16}N_2O$) C, H, N.

(*Z*)-3-[(3-Bromothien-2-yl)methylidenyl]indolin-2-one (54): ^{1}H NMR (300 MHz, DMSO-d_6) δ 10.72 (s, 1H, NH-1), 8.00 (d, J = 5.62 Hz, 1H, H-4′(5′)), 7.82 (s, 1H, H-vinyl), 7.65 (d, J = 7.61 Hz, 1H, H-4), 7.35 (d, J = 5.62 Hz, 1H, H-5′-(4′)), 7.26 (dt, J = 1.03, 7.61 Hz, 1H, H-6), 7.02 (dt, J = 0.84, 7.61 Hz, 1H, H-5), 6.88 (d, J = 7.61 Hz, 1H, H-7); MS m/z (relative intensity, %) 306 (43, $M^{+\bullet}$). Anal. ($C_{13}H_8BrNOS$) C, H, N.

(*Z*)-3-[(3-Bromothien-5-yl)methylidenyl]indolin-2-one (55): ^{1}H NMR (300 MHz, DMSO-d_6) δ 10.70 (s, 1H, NH-1), 8.04 (s, 1H, H-vinyl), 7.95−7.97 (m, 2H, H-2′,4′), 7.59 (d, J = 7.57 Hz, 1H, H-4), 7.23 (dt, J = 1.10, 7.57 Hz, 1H, H-6), 7.01 (dt, J = 0.87, 7.57 Hz, 1H, H-5), 6.87 (d, J = 7.57 Hz, 1H, H-7); MS m/z (relative intensity, %) 306 (100, $M^{+\bullet}$). Anal. ($C_{13}H_8BrNOS$) C, H, N.

(*Z*)-3-[(2-(Methylthio)thien-5-yl)methylidenyl]indolin-2-one (56). A reaction mixture of 134.0 mg of oxindole, 189.9 mg of the 5-(methylthio)thiophene-2-carboxaldehyde, and 3 drops of piperidine in 2 mL of ethanol was stirred at 90 °C for 3 h. After cooling, the precipitate was filtered, washed with cold ethanol, and dried to yield 246.6 mg (90%) of the title compound as an orange solid: ^{1}H NMR (360 MHz, DMSO-d_6) δ 10.56 (s, br, 1H, NH-1), 7.98 (s, 1H, H-vinyl), 7.73 (d, J = 4.28 Hz, 1H, H-4′(3′)), 7.64 (d, J = 7.54 Hz, 1H, H-4), 7.19 (dt, J = 1.11, 7.54 Hz, 1H, H-6), 7.13 (d, J = 4.28 Hz, 1H, H-3′-(4′)), 6.98 (dt, J = 0.75, 7.54 Hz, 1H, H-5), 6.85 (d, J = 7.54 Hz, 1H, H-7), 2.64 (s, 3H, SCH_3-2′); MS m/z (relative intensity, %) 274 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{14}H_{11}NOS_2$) C, H, N.

(*Z*)-4-Methyl-3-[(2-(methylthio)thien-5-yl)methylidenyl]-indolin-2-one (57): ^{1}H NMR (300 MHz, DMSO-d_6) δ 10.60 (s, br, NH-1), 7.87 (s, 1H, H-vinyl), 7.76 (d, J = 4.33 Hz, 1H, H-4′), 7.12 (d, J = 4.33 Hz, 1H, H-3′), 7.08 (t, J = 7.92 Hz, 1H, H-6), 6.78 (d, J = 7.92 Hz, 1H, H-5), 6.72 (d, J = 7.92 Hz, 1H, H-7), 2.63 (s, 3H, SCH_3-2′), 2.57 (s, 3H, CH_3-4); 2D-NOE ^{1}H NMR showed the correlation between H-vinyl and CH_3-4, which confirmed the *Z* conformer; MS m/z (relative intensity, %) 310 (100, $[M + Na]^{+\bullet}$). Anal. ($C_{15}H_{13}NOS_2 \cdot 0.1H_2O$) C, H, N.

(*E*)-3-[(2-(Ethylthio)thien-5-yl)methylidenyl]indolin-2-one (58): ^{1}H NMR (360 MHz, DMSO-d_6) δ 10.52 (s, 1H, NH-1), 7.98 (s, 1H, H-vinyl), 7.76 (d, J = 3.92 Hz, 1H, H-4′(3′)), 7.64 (d, J = 7.40 Hz, 1H, H-4), 7.17 (dt, J = 1.13, 7.40 Hz, 1H, H-6), 6.95−6.99 (m, 2H, H-5,3′(4′)), 6.84 (d, J = 7.40 Hz, 1H, H-7), 2.90 (q, J = 7.49 Hz, 2H, C*H*$_2$CH$_3$-2), and 1.29 (t, J = 7.49 Hz, 3H, CH_2C*H*$_3$-2); MS m/z (relative intensity, %) 256 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{15}H_{13}NOS$) C, H, N.

(*E*)-3-[(Furan-2-yl)methylidenyl]indolin-2-one (59): ^{1}H NMR (360 MHz, DMSO-d_6) δ 10.58 (s, 1H, NH-1), 8.35 (d, J = 7.55 Hz, 1H, H-4), 8.13 (d, J = 1.87 Hz, 1H, H-5′), 7.33 (s, 1H, H-vinyl), 7.22−7.26 (m, 2H, H-6,3′), 7.00 (dt, J = 1.08, 7.55 Hz, 1H, H-5), 6.87 (d, J = 7.59 Hz, 1H, H-7), 6.78 (dd, J = 1.87, 3.11 Hz, H-4′); MS m/z (relative intensity, %) 212 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{13}H_9NO_2$) C, H, N.

(*E*)-3-[(2-Methylfuran-5-yl)methylidenyl]indolin-2-one (60): ^{1}H NMR (360 MHz, DMSO-d_6) δ 10.48 (s, 1H, NH-1), 8.29 (d, J = 7.78 Hz, 1H, H-4), 7.24 (s, 1H, H-vinyl), 7.22 (t, J = 7.67 Hz, 1H, H-6), 7.15 (d, J = 3.28 Hz, 1H, H-4′), 7.01 (t, J = 7.78 Hz, 1H, H-5), 6.86 (d, J = 7.67 Hz, 1H, H-7), 6.44 (d, J = 3.28 Hz, 1H, H-3′), 2.51 (s, 3H, CH_3-2′); MS m/z (relative intensity, %) 226 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{14}H_{11}NO_2$) C, H, N.

(*E*)-3-[(2-Ethylfuran-5-yl)methylidenyl]indolin-2-one (61): ^{1}H NMR (360 MHz, DMSO-d_6) δ 10.49 (s, 1H, NH-1), 8.31 (d, J = 7.68 Hz, 1H, H-4), 7.25 (s, 1H, H-vinyl), 7.21 (dt, J = 1.08, 7.60 Hz, 1H, H-6), 7.15 (d, J = 3.16 Hz, 1H, H-4′), 7.00 (dt, J = 1.08, 7.68 Hz, 1H, H-5), 6.86 (d, J = 7.60 Hz, 1H, H-7), 6.45 (d, J = 3.16 Hz, 1H, H-3′), 2.85 (q, J = 7.49 Hz, 2H, C*H*$_2$CH$_3$-2′), 1.29 (t, J = 7.49 Hz, 3H, CH_2C*H*$_3$-2′); MS m/z (relative intensity, %) 240 (100, $[M + 1]^{+\bullet}$). Anal. ($C_{15}H_{13}NO_2$) C, H, N.

(*Z*)-3-[(4-Chloropyrazol-3-yl)methylidenyl]indolin-2-one (62): ^{1}H NMR (360 MHz, DMSO-d_6) δ 14.59 (s, 1H, NH-1′), 11.26 (s, 1H, br, NH-1), 7.86−7.88 (m, 2H, H-4,5′), 7.53 (s, 1H, H-vinyl), 7.30 (dt, J = 1.03, 7.74 Hz, 1H, H-6), 7.07 (dt, J = 1.02, 7.74 Hz, 1H, H-5), and 6.94 (d, J = 7.74 Hz, 1H, H-7); MS m/z (relative intensity, %) 246 (100, $M^{+\bullet}$). Anal. ($C_{12}H_8ClN_3O$) C, H, N.

(*E*)-3-[(4-Chloro-1-methylpyrazol-3-yl)methylidenyl]-indolin-2-one (63): ^{1}H NMR (360 MHz, DMSO-d_6) δ 10.57 (s, 1H, NH-1), 8.88 (d, J = 7.57 Hz, 1H, H-4), 8.19 (s, 1H, H-vinyl), 7.34 (s, 1H, H-5′), 7.28 (dt, J = 1.14, 7.57 Hz, 1H, H-6), 7.02 (dt, J = 1.07, 7.57 Hz, 1H, H-5), 6.87 (d, J = 7.57 Hz, 1H, H-7), 4.05 (s, 3H, CH_3-1′); MS m/z (relative intensity, %) 260 (100, $M^{+\bullet}$). Anal. ($C_{13}H_{10}ClN_3O$) C, H, N.

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(20) Compound **45** has been named as either 3-[(2,4-dimethylpyrrol-5-yl)methylidenyl]indolin-2-one or 3-[(3,5-dimethylpyrrol-2-yl)-methylidenyl]indolin-2-one in this paper and other presentations. However, the wrong name, 3-[(2,3-dimethylpyrrol-5-yl)methylidenyl]indolin-2-one, was given in our patent (PCT Int. Appl. 293pp, WO 9807695).

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