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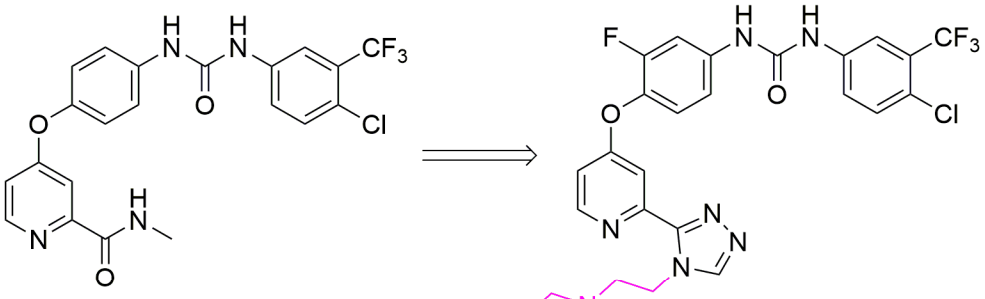

Sorafenib **13i**

ClogD$_{7.4}$ = 4.34
IC$_{50}$ HT-29 cells 3.37 uM
H460 cells 2.25 uM
MDA-MB-231 cells 3.08 uM
WI-38 normal cells 8.42 uM

ClogD$_{7.4}$ = 3.18
IC$_{50}$ HT-29 cells 0.90 uM
H460 cells 0.85 uM
MDA-MB-231 cells 1.54 uM
WI-38 normal cells 6.73 uM

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Discovery of novel diaryl urea derivatives bearing a triazole moiety as potential antitumor agents

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Abstract

Herein, we report a novel series of diaryl urea derivatives bearing a triazole moiety, from which potent antitumor agents have been identified. With a modified triazole, most compounds showed high level activity in both cellular and enzymatic assays, accompanied with a suitable $ClogD_{7.4}$ value. The most active compound, **13i**, effectively suppressed proliferation of HT-29, H460 and MDA-MB-231 cancer cells, with IC_{50} values of 0.90, 0.85 and 1.54 μM, respectively. Compound **13i** also exhibited significant inhibition of tyrosine kinases including c-Kit, RET and FLT3. Furthermore, compound **13i** could obviously induce apoptosis of HT-29 cells in a concentration-dependent manner. The study of structure-activity relationships also revealed that a hydrophilic tail at the 4-position of the triazole was crucial for high activity of the compound.

Keywords: Diaryl ureas; Organic synthesis; Antitumor activity; Structure-activity relationship

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1. Introduction

Cancer is a serious worldwide public health concern. Most patients suffering from cancer have a poor prognosis due to its high mortality rate and incidence of relapse. Therefore, there is still an urgent need for new antitumor agents with improved efficacy.

In the past two decades, multikinase inhibitors, such as sunitinib, imatinib, and sorafenib have received great attention and have yielded great benefits in numerous clinical cancer cases [1–4]. The simultaneous inhibition of multiple targets can create a synergistic anti-cancer effect. This strategy is proven to efficiently interrupt complex oncogenic pathways and reduce the possibility of developing drug resistance [5]. Sorafenib, a well-known multikinase inhibitor, potently suppresses several receptor tyrosine kinases, including PDGFR-β, VEGFR-2, VEGFR-3, FLT3, Kit, and RET, as well as the downstream protein Raf. In addition, it demonstrates significant inhibition across a broad spectrum of tumour types [6–8]. Thus far, it has been approved by the U.S. Food and Drug Administration (FDA) for the treatment of advanced renal carcinoma (RCC), unresectable hepatocellular carcinoma (HCC), and differentiated thyroid carcinoma (DTC). In addition, its applications in breast, lung, and colorectal carcinomas are being evaluated in clinic, and the results are eagerly awaited [9–12].

Despite its outstanding antitumor activity, the overall bioavailability of sorafenib is limited, mainly because of its poor water-solubility which is ascribed to the high hydrophobicity of its chemical structure [13,14]. We considered rational optimization of sorafenib a feasible approach to develop novel antitumor agents, placing equal attention and importance on their antitumor potency as well as their hydrophilicity.

As reported previously, the diaryl urea framework plays a pivotal role in the complexation of sorafenib with enzymes through hydrogen bonding and hydrophobic interactions [15], which prompted us to retain this functional template in the design of new compounds. In previous study, we have reported a series of compounds based on a 2-(4-(2-(dimethylamino)ethyl)-4*H*-1,2,4-triazol-3-yl)pyridine moiety as potent antitumor agents [16]. Structure-activity relationships revealed that an optimized triazole was crucial for high activity. Thus, in the current research, we developed a strategy to investigate hybrids of diaryl urea and triazole for application as antitumor agents.

Here, we reported the design, synthesis and biological evaluation of a series of diaryl urea derivatives bearing a triazole moiety, to identify their potential as antitumor agents (Fig. 1). The new compounds were screened in HT-29, H460, and MDA-MB-231 cancer cells, and against a kinase panel comprising c-Kit, RET, FLT3, B-Raf, and VEGFR-2. Additionally, cell apoptosis induced by compound **13i** was evaluated in HT-29 cells.

(Insert **Fig. 1** here)

2. Results and discussion

2.1 Chemistry

The general route to prepare compounds **8a** and **8b** is described in Scheme 1. The appropriate aniline was reacted with triphosgene in refluxing toluene to give the corresponding isocyanate **1a–f** as a solid or an oil. The synthesis of intermediate **2** was achieved in two steps from picolinic acid, which has been described in detail in our previous study [16]. A nucleophilic substitution of **2** with 2-fluoro-4-nitrophenol in refluxing chlorobenzene yielded the crude product of **3**, which was purified by recrystallization in absolute ethanol. Aminolysis of **3** with ammonia in acetone led to the desired intermediate **4**, which then reacted with *N*,*N*-dimethylformamide dimethyl acetal (DMF-DMA) to generate **5** in a high yield. Intermediate **6** was obtained by cyclization of **5** with methylhydrazine under a modified condition, as previously reported [17] .Then, intermediate **6** was reduced using hydrazine hydrate, to efficiently yield amine **7**. Ureas **8a** and **8b** were obtained by reaction of intermediate **7** with the corresponding isocyanate.

(Insert **Scheme** 1 here)

The synthetic procedure for compounds **13a–y** is illustrated in Scheme 2. As shown, these target compounds were synthesized via a five-step reaction from intermediate **3**. The amino intermediate **9** was afforded by the reduction of intermediate **3** in the presence of Pd/C, and was then reacted with the appropriate isocyanates to generate urea intermediates **10a–f**. A convenient hydrazinolysis of **10a–f** in MeCN resulted in **11a–f**, respectively. The condensation of **11a–f** with DMF-DMA was accomplished under similar conditions as described for intermediate **5**, and generated **12a–f**, respectively. The proposed compounds **13a–y** were generated by cyclization of **12a–f** with the appropriate amines in the presence of acetic acid [16]. All the target compounds were purified by column chromatography on silica gel. The chemical structures of the target compounds were confirmed by elemental analysis, mass spectrometry and NMR.

(Insert **Scheme 2** here)

2.2. Biological evaluation

2.2.1. In vitro cellular activity and structure-activity relationships

To get a preliminary insight into SARs, our strategy was to start by testing compounds **8a**, **8b**, **13a**, and **13b**, which shared a diaryl urea template but were distinct in the arrangement of the triazole and distal aryl moieties. The antitumor activity of these compounds was screened in three cancer cell lines, namely HT-29 (human colon cancer), H460 (human lung cancer), and MDA-MB-231 (human breast cancer), as well as in a non-malignant cell line WI-38 (human fetal lung fibroblasts), using standard MTT assays. Sorafenib was used as a positive control. The biological data, expressed as IC_{50} values, are listed in Table 1.

(Insert **Table 1** here)

Interestingly, the results of this screening revealed dramatic differences in the activities of these compounds. Both compounds **8a** and **8b**, which possessed a methyl group on 1-position of the triazole, were practically inactive against all the tested cancer cell lines. However, shifting the methyl to 4-position led to a significant improvement in potency, as shown with compounds **13a** and **13b**. The results indicated a clear SAR for the triazole moiety, as a suitable substitution on 4-position was crucial for high potency. On the other hand, the antitumor activity of the compounds was only fine-tuned by the pattern on the distal aryl fragment. Compound **13b** with a CF_3 group at meta-position possessed moderate activity, having an IC_{50} value of 3.02 μM against HT-29 cells, and 1.27 μM against H460 cells. Compound **13a**, which contained an additional chlorine atom (R_2 = 3-CF_3-4-Cl), exhibited a slightly increased inhibition against HT-29 cells compared to **13b**, but was less potent in H460 cells. Given these encouraging results, a more detailed investigation of SARs was performed with compounds substituted on the 4-position of the triazole. Meanwhile, to ameliorate the physicochemical properties of the new compounds, diverse moieties with different hydrophilicities were introduced. In addition, the distal aryl fragment was optimized with the aim of further promoting the compounds' potencies. Accordingly, compounds **13c–y** were prepared, and assessed for activity in HT-29, H460, MDA-MB-231, and WI-38 cell lines (Table 2).

(Insert **Table 2** here)

The newly synthesized compounds exhibited significant inhibition of cancer cell proliferation. Compounds **13a–d**, **13g–j**, and **13l–u** were more potent than sorafenib against one or more of the tested cancer cell lines. Among them, compounds **13c** and **13i** effectively suppressed proliferation of all the tested cancer cell lines, with IC_{50} values between 0.88–1.15 and 0.85–1.54 μM, respectively. On the other hand, most of the tested compounds were less toxic to human fibroblasts WI-38 in comparison to their effects on cancer cells, which indicated a favorable safety of the compounds.

Further SAR investigations into modifications on the 4-position of the triazole revealed that a hydrophilic tail yielded the desired biological activity and physicochemical properties. Compound **13a**, which possessed a methyl group on the triazole exhibited modest activity against HT-29 cells (IC_{50}, 2.57 μM). However, substitution for a cyclopropyl group, providing compound **13y**, sharply reduced potency (IC_{50}, 10.90 μM). In contrast, an evident improvement in inhibitory activity was observed when a hydrophilic tail was incorporated, as shown by compounds **13c**, **13i**, and **13o** (IC_{50}: 0.88, 0.90, and 1.40 μM, respectively). Compounds **13c** and **13i** showed good activities against MDA-MB-231 cells (IC_{50}: 1.07 and 1.54 μM, respectively), but compound **13o** was less effective (IC_{50}: 7.88 μM), which indicated that the length of chain might be related to cellular selectivity. It was noted that with a morpholinyl group, compound **13u**, showed an unfavorable overall potency (IC_{50}, 2.86–8.28 μM), indicating that bulky moieties were not effective. Also noteworthy is that

all the compounds bearing a hydrophilic tail possessed an attractive $ClogD_{7.4}$, which suggests a good drug distribution in blood serum.

The results regarding modifications of the distal aryl group revealed that placing a CF_3 group in the meta-position was most suitable. Compound **13c** (R_2 = 3-CF_3-4-Cl) and **13d** (R_2 = 3-CF_3) potently inhibited proliferation of HT-29 and H460 cells, showing IC_{50} values between 0.88–2.15 μM. However, decreased activity was observed for compounds **13f–h**, which held a F or Cl atom in R_2. Moreover, shifting the CF_3 group to the ortho-position (**13e**) resulted in a complete loss of inhibition in H460 cells, as well as a slight decrease in the inhibition of HT-29 cells (IC_{50}, 5.86 μM). Similar results were observed when **13i** and **13j** were compared with **13k–n**.

Regarding MDA-MB-231 cells, compound **13c**, which bore a 3-CF_3-4-Cl group, displayed a high level of activity, exhibiting an IC_{50} value of 1.07 μM. However, any change of R_2 led to a significant decline in potency. For example, compounds **13d** (R_2 = 3-CF_3), **13g** (R_2 = 3-Cl-4-F) and **13h** (R_2 = 2-4-$(Cl)_2$) exhibited only week inhibition of MDA-MB-231 cells (IC_{50}, 16.35, 8.32 and 7.96 μM, respectively), while compounds **13e** (R_2 = 2-CF_3) and **13f** (R_2 = 3,4-$(F)_2$) were totally inactive. Overall, the 3-CF_3-4-Cl group was evaluated as the optimal configuration for R_2, which is consistent with the framework of sorafenib.

2.2.2. In vitro enzymatic assay

To rationalize their appealing cell growth inhibition, compounds **13c–u** and **13x** were screened against a kinase panel comprising c-Kit, RET, FLT3, B-Raf and VEGFR-2. The enzymatic experiments were established using a mobility shift assay, the results are summarized in Table 3.

(Insert **Table 3** here)

It is interesting that several compounds (**13c**, **13d**, **13g**, **13i**, **13j**, **13m**, **13o**, **13p**, **13r**, **13s**, and **13u**) exhibited significant inhibition of the tested kinases at a concentration of 10 μM. Compounds **13c**, **13g**, **13i**, **13j**, **13o** and **13p** were evaluated as triple kinase inhibitors of c-Kit, RET and FLT3. Compound **13d** potently inhibited c-Kit and RET, while compound **13s** effectively suppressed c-Kit and FLT3. On the other hand, all the tested compounds showed poor potency against B-Raf and VEGFR-2 except for **13p**, indicating that these compounds might have a different mechanism of action with sorafenib. Further studies into SARs revealed that placing a 3-CF_3-4-Cl group at R_2 produced the best kinase inhibition, which is consistent with the results obtained in the cellular assay.

The optimal enzyme inhibition was exhibited by compound **13i**, which inhibited multiple kinases, c-Kit, RET and FLT3, by approximately 80%. Nonetheless, it was still less active than sorafenib, which suggests that the selected kinases may not be the precise biological targets of these new compounds. Detailed investigations on their mechanisms of action are underway.

2.2.3. Cell apoptosis analysis

Cell apoptosis is the process of programmed cell death, and induction of apoptosis is considered a major way that most antitumor drugs work [18]. To determine the mode of cell death induced by compound **13i**, a biparametric cytofluorimetric analysis was performed using propidium iodide (PI) and annexinV-FITC staining in HT-29 cells. After treatment with compound **13i** for 48 h at different concentrations (0, 1, 2, 5 μM) , the stained cells were analyzed by a flow cytometer. As shown in Figure 2, the apoptotic rate of HT-29 cells was obviously increased from 4.21% (control) to 48.10%, which suggested that compound **13i** caused a significant induction of cell apoptosis in a concentration-dependent manner.

(Insert **Fig. 2** here)

3. Conclusions

In summary, interesting diaryl ureas bearing a modified triazole have been identified as potent antitumor agents. We provided a valuable rational design strategy that aimed to improve both the antitumor activity as well as the physicochemical properties of sorafenib. Detailed SAR studies on the triazole and distal phenyl ring were undertaken, the efforts of which led to the discovery of an optimal compound, **13i**, which displayed high activity in both cellular and enzymatic assays. Flow cytometry analysis demonstrated that compound **13i** inhibited the proliferation of HT-29 cells by inducing apoptosis. In addition, compound **13i** had a favourable $ClogD_{7.4}$ value, which is crucial for its in vivo bioavailability and activity. Taken together, compound **13i** is a promising lead compound for future antitumor drug development. Further investigations into its pharmacokinetics and *in vivo* activity are in progress, the results will be reported in due course.

4. Experimental section

4.1. Chemistry

Reagents and solvents were obtained from commercial sources and used without further purification. The purity of the synthesized compounds was measured by high performance liquid chromatography (HPLC, Agilent, USA). Flash chromatography was performed using silica gel (300–400 mesh). All reactions were monitored by TLC on silica gel plates. Melting points were determined on a Büchi Melting Point B-540 apparatus (Büchi Labortechnik, Flawil, Switzerland). The ^{1}H and ^{13}C NMR were recorded on Bruker ARX-400 or ARX-600 spectrometer (Bruker Bioscience, Billerica, MA, USA) with TMS as an internal standard. Mass spectra (MS) were measured in ESI mode on an Agilent 1100 LC-MS (Agilent, Palo Alto, CA, USA). Elemental analysis was determined on a Carlo-Erba 1106 Elemental analysis instrument (Carlo Erba, Milan, Italy).

*4.1.1. General procedure for preparation of isocyanates (**1a–f**)*

To a stirred and cooled to 0 °C solution of triphosgene (0.1 mol) in toluene, appropriate anilines (0.3 mol) was added slowly. The mixture was stirred for 0.5 h at room temperature, and for another 7–10 h under reflux until TLC showed the completion of the reaction. The solvent was evaporated, the mixture was distilled under reduced pressure to afford intermediates **1a–f**.

*4.1.1.1. 1-Chloro-4-isocyanato-2-(trifluoromethyl)benzene (**1a**).* White solid; 150–151 °C (15 mmHg).

*4.1.1.2. 1-Isocyanato-3-(trifluoromethyl)benzene (**1b**).* White solid; 172–174 °C (15 mmHg).

*4.1.1.3. 1-Isocyanato-2-(trifluoromethyl)benzene (**1c**).* Colorless oil; 165–167°C (15 mmHg).

*4.1.1.4. 1,2-Difluoro-4-isocyanatobenzene (**1d**).* Colorless oil; 177–179°C (15 mmHg).

*4.1.1.5. 2-Chloro-1-fluoro-4-isocyanatobenzene (**1e**).* Colorless oil; 159–161°C (15 mmHg).

*4.1.1.6. 2,4-Dichloro-1-isocyanatobenzene (**1f**).* Colorless oil; 183–185°C (15 mmHg).

*4.1.2. 3-Fluoro-4-(2-(1-methyl-1H-1,2,4-triazol-5-yl)pyridin-4-yloxy)aniline (**7**).*

The synthesis of intermediate **7** has been described in detail in our previous work [16], so the synthetic method is not listed here. Gray solid; Yield: 63%; M.p. 150–153 °C; ^{1}H NMR (300 MHz, DMSO-d_6) δ 8.61 (d, J = 5.7 Hz, 1H, ArH), 7.99 (s, 1H, ArH), 7.48 (d, J = 2.4 Hz, 1H, ArH), 7.01–7.14 (m, 2H, ArH), 6.56 (dd, J = 13.2, 2.4 Hz, 1H, ArH), 6.47 (dd, J = 8.7, 2.1 Hz, 1H, ArH), 5.56 (s, 2H, NH), 4.27 (s, 3H, CH_3); ESI-MS m/z: 285.8 $[M+H]^+$.

*4.1.3. General procedure for preparation of the target compounds **8a** and **8b***

At room temperature, to a stirred solution of intermediate **7** (0.7 mmol) in CH_2Cl_2, **1a–b** (0.77 mmol) was added. The mixture was stirred for 5 h and monitored by TLC. The precipitate was collected by filtration, washed with Et_2O, and dried to give the solids **8a** and **8b**, respectively.

4.1.3.1.
*1-(4-Chloro-3-(trifluoromethyl)phenyl)-3-(3-fluoro-4-(2-(1-methyl-1H-1,2,4-triazol-5-yl)pyridin-4-yloxy)phenyl)urea (**8a**).* HPLC purity: 98.2%; White solid; Yield: 68%; M.p. 218.9–219.8 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.35 (s, 1H, NH), 9.26 (s, 1H, NH), 8.65 (d, J = 5.7 Hz, 1H, ArH), 8.12 (s, 1H, ArH), 7.99 (s, 1H, ArH), 7.74 (d, J = 12.8 Hz, 1H, ArH), 7.62–7.70 (m, 2H, ArH), 7.52 (d, J = 2.2 Hz, 1H, ArH), 7.40 (t, J = 8.9 Hz, 1H, ArH), 7.31 (d, J = 8.4 Hz, 1H, ArH), 7.17 (dd, J = 5.6, 2.5 Hz, 1H, ArH), 4.27 (s, 3H, CH_3); ^{13}C NMR (101 MHz, DMSO-d_6) δ 165.39, 153.98, 152.83, 151.80, 151.07, 150.53, 149.87, 139.56, 139.22, 134.38, 132.48, 127.19, 124.42, 123.77, 123.26, 123.09, 117.46, 115.92, 112.41, 109.66, 107.82, 39.04; ESI-MS m/z: 507.1 $[M+H]^+$; Anal. calcd. for $C_{22}H_{15}ClF_4N_6O_2$ (%): C, 52.13; H, 2.98; N, 16.58. Found (%): C, 52.19; H, 3.07; N, 16.65.

4.1.3.2.
*1-(3-Fluoro-4-(2-(1-methyl-1H-1,2,4-triazol-5-yl)pyridin-4-yloxy)phenyl)-3-(3-(trifluoromethyl)phenyl)urea (**8b**).* HPLC purity: 98.7%; White solid; Yield:

72%; M.p. 206.8–209.4 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.22 (s, 1H, NH), 9.20 (s, 1H, NH), 8.65 (d, *J* = 5.5 Hz, 1H, ArH), 8.03 (s, 1H, ArH), 7.99 (s, 1H, ArH), 7.76 (d, *J* = 13.0 Hz, 1H, ArH), 7.62 (d, *J* = 6.7 Hz, 1H), 7.52–7.56 (m, 2H, ArH), 7.32–7.42 (m, 3H, ArH), 7.18 (d, *J* = 3.3 Hz, 1H, ArH), 4.27 (s, 3H, CH_3); ^{13}C NMR (101 MHz, DMSO-d_6) δ 165.42, 154.00, 152.91, 151.81, 151.08, 150.53, 149.87, 140.76, 139.34, 134.28, 130.43, 130.01, 124.66, 124.44, 122.57, 118.87, 115.79, 114.84, 112.43, 109.65, 107.70, 39.04; ESI-MS m/z: 473.1 $[M+H]^+$; Anal. calcd. for $C_{22}H_{16}F_4N_6O_2$ (%): C, 55.94; H, 3.41; N, 17.79. Found (%): C, 55.86; H, 3.37; N, 18.13.

*4.1.4. Methyl 4-(4-amino-2-fluorophenoxy)picolinate (**9**)*

To a solution of intermediate **3** (5 g, 0.017 mol) in ethanol, 0.5 g palladium on charcoal (10%) was added. The mixture was stirred for 4 h at room temperature. After that time, the catalyst was filtered off and the filtrate was evaporated. The residue was triturated with diethyl ether, filtered off, and dried to give intermediate **9** as a pale-yellow solid (2.72 g, 60.6%); M.p. 69.5–71.5 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.54 (dd, *J* = 5.5, 3.2 Hz, 1H, ArH), 7.39 (dd, *J* = 9.5, 2.5 Hz, 1H, ArH), 7.14 (ddd, *J* = 12.7, 5.6, 2.6 Hz, 1H, ArH), 7.02 (t, *J* = 9.0 Hz, 1H, ArH), 6.51 (dd, *J* = 13.2, 2.5 Hz, 1H, ArH), 6.43 (dd, *J* = 8.7, 2.1 Hz, 1H, ArH), 5.52 (s, 2H, NH_2), 3.83 (s, 3H, CH_3); ESI-MS m/z: 263.0 $[M+H]^+$.

*4.1.5. General procedure for preparation of intermediates **10a–f***

A mixture of intermediate **9** (2 mmol) and appropriate isocyanate (**1a–e**, 2 mmol) in CH_2Cl_2 was stirred at room temperature for 4–7 h until TLC showed the completion of reaction. The precipitate was filtered off and washed to give the solids **10a–f**.

*4.1.5.1. Methyl 4-(4-(3-(4-chloro-3-(trifluoromethyl)phenyl)ureido)-2-fluorophenoxy)picolinate (**10a**).* White solid; Yield: 92%; M.p. 133.6–134.5 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.35 (s, 1H, NH), 9.27 (s, 1H, NH), 8.59 (d, *J* = 5.6 Hz, 1H, ArH), 8.09 (d, *J* = 2.1 Hz, 1H, ArH), 7.71 (dd, *J* = 13.2, 2.2 Hz, 1H, ArH), 7.66 (dd, *J* = 8.9, 2.2 Hz, 1H, ArH), 7.62 (d, *J* = 8.8 Hz, 1H, ArH), 7.45 (dd, *J* = 7.4, 2.5 Hz, 1H, ArH), 7.36 (t, *J* = 8.9 Hz, 1H, ArH), 7.28 (dd, *J* = 8.9, 1.7 Hz, 1H, ArH), 7.22 (dd, *J* = 5.6, 2.6 Hz, 1H, ArH), 3.84 (s, 3H, CH_3); ESI-MS m/z: 484.5 $[M+H]^+$.

*4.1.5.2. Methyl 4-(2-fluoro-4-(3-(3-(trifluoromethyl)phenyl)ureido)phenoxy)picolinate (**10b**).* White solid; Yield: 86%; M.p. 169.7–171.1 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.20 (s, 1H, NH), 9.18 (s, 1H, NH), 8.59 (d, *J* = 5.6 Hz, 1H, ArH), 8.00 (s, 1H, ArH), 7.73 (dd, *J* = 13.2, 2.4 Hz, 1H, ArH), 7.59 (d, *J* = 8.6 Hz, 1H, ArH), 7.52 (t, *J* = 7.9 Hz, 1H, ArH), 7.45 (dd, *J* = 6.9, 2.6 Hz, 1H, ArH), 7.31–7.39 (m, 2H, ArH), 7.27 (dd, *J* = 8.9, 1.7 Hz, 1H, ArH), 7.22 (dd, *J* = 5.6, 2.6 Hz, 1H, ArH), 3.84 (s, 3H, CH_3); ESI-MS m/z: 472.1 $[M+Na]^+$.

*4.1.5.3. Methyl 4-(2-fluoro-4-(3-(2-(trifluoromethyl)phenyl)ureido)phenoxy)picolinate (**10c**).* White solid; Yield: 81%; M.p. 122.2–125.1 °C; ^{1}H NMR (400 MHz,

DMSO-d_6) δ 9.68 (s, 1H, NH), 8.58 (dd, *J* = 5.6, 2.3 Hz, 1H, ArH), 8.19 (s, 1H, NH), 7.89 (d, *J* = 8.2 Hz, 1H, ArH), 7.74 (dd, *J* = 13.3, 2.4 Hz, 1H, ArH), 7.62–7.71 (m, 2H, ArH), 7.45 (dd, *J* = 7.7, 2.5 Hz, 1H, ArH), 7.33 (dt, *J* = 15.5, 8.3 Hz, 2H, ArH), 7.21 (ddd, *J* = 8.5, 5.3, 2.4 Hz, 2H, ArH), 3.84 (s, 3H, CH_3); ESI-MS m/z: 472.2 $[M+Na]^+$.

*4.1.5.4. Methyl 4-(4-(3-(3,4-difluorophenyl)ureido)-2-fluorophenoxy)picolinate (**10d**).* White solid; Yield: 86%; M.p. 213.0–214.7 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.13 (s, 1H, NH), 9.04 (s, 1H, NH), 8.59 (d, *J* = 5.6 Hz, 1H, ArH), 7.70 (dd, *J* = 13.3, 2.3 Hz, 1H, ArH), 7.65 (ddd, *J* = 13.3, 7.5, 2.6 Hz, 1H, ArH), 7.43 (d, *J* = 2.3 Hz, 1H, ArH), 7.31–7.39 (m, 2H, ArH), 7.25 (dd, *J* = 9.1, 1.3 Hz, 1H, ArH), 7.21 (dd, *J* = 5.6, 2.5 Hz, 1H, ArH), 7.12–7.16 (m, 1H, ArH), 3.83 (s, 3H, CH_3); ESI-MS m/z: 418.1 $[M+H]^+$.

*4.1.5.5. Methyl 4-(4-(3-(3-chloro-4-fluorophenyl)ureido)-2-fluorophenoxy)picolinate (**10e**).* White solid; Yield: 90%; M.p. 213.6–215.4 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.15 (s, 1H, NH), 9.03 (s, 1H, NH), 8.58 (d, *J* = 5.6 Hz, 1H, ArH), 7.78 (dd, *J* = 6.7, 1.6 Hz, 1H, ArH), 7.71 (dd, *J* = 13.2, 2.4 Hz, 1H, ArH), 7.44 (dd, *J* = 7.1, 2.5 Hz, 1H, ArH), 7.30–7.38 (m, 3H, ArH), 7.25 (dd, *J* = 8.9, 1.5 Hz, 1H, ArH), 7.21 (dd, *J* = 5.6, 2.6 Hz, 1H, ArH), 3.83 (s, 3H, CH_3); ESI-MS m/z: 434.0 $[M+H]^+$.

*4.1.5.6. Methyl 4-(4-(3-(2,4-dichlorophenyl)ureido)-2-fluorophenoxy)picolinate (**10f**).* White solid; Yield: 77%; M.p. 195.5–198.0 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.80 (s, 1H, NH), 8.61 (d, *J* = 5.6 Hz, 1H, ArH), 8.52 (s, 1H, NH), 8.17 (d, *J* = 9.0 Hz, 1H, ArH), 7.75 (dd, *J* = 13.1, 2.3 Hz, 1H, ArH), 7.66 (d, *J* = 2.4 Hz, 1H, ArH), 7.47 (dd, *J* = 7.2, 2.5 Hz, 1H, ArH), 7.37–7.43 (m, 2H, ArH), 7.24 (dd, *J* = 5.7, 2.4 Hz, 2H, ArH), 3.86 (s, 3H, CH_3); ESI-MS m/z: 450.1 $[M+H]^+$.

*4.1.6. General procedure for preparation of intermediates **11a–f***

To a solution of **10a–f** (1 mmol) in MeCN, 80% hydrazine hydrate (3 mmol) was added dropwise. The mixture was sirred under refulx and monitored by TLC. The solvent was concentrated and cooled to room temperature. The precipitate was collected by filtration and washed to yield the corresponding compounds **11a–f**.

*4.1.6.1. 1-(4-Chloro-3-(trifluoromethyl)phenyl)-3-(3-fluoro-4-((2-(hydrazinecarbonyl)pyridin-4-yl)oxy)phenyl)urea (**11a**).* White solid; Yield: 72%; M.p. 173.5–175.0 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.92 (s, 1H, –N<u>H</u>–NH_2), 9.37 (s, 1H, NH), 9.27 (s, 1H, NH), 8.49 (br, 1H, ArH), 8.10 (s, 1H, ArH), 7.71 (d, *J* = 12.4 Hz, 1H, ArH), 7.64 (dd, *J* = 18.2, 9.5 Hz, 2H, ArH), 7.36 (m, 2H, ArH), 7.28 (dd, *J* = 7.9, 0.8 Hz, 1H, ArH), 7.16 (d, *J* = 2.4 Hz, 1H, ArH), 4.54 (s, 2H, –NH–N<u>H</u>$_2$). ESI-MS m/z: 506.4 $[M+Na]^+$.

*4.1.6.2. 1-(3-Fluoro-4-((2-(hydrazinecarbonyl)pyridin-4-yl)oxy)phenyl)-3-(3-(trifluoromethyl)phenyl)urea (**11b**).* White solid; Yield: 69%; M.p. 183.4–185.7 °C; ^{1}H

NMR (400 MHz, DMSO-d_6) δ 9.97 (s, 1H, –N<u>H</u>–NH_2), 9.22 (s, 1H, NH), 9.19 (s, 1H, NH), 8.52 (d, *J* = 5.6 Hz, 1H, ArH), 8.02 (s, 1H, ArH), 7.75 (dd, *J* = 13.1, 2.3 Hz, 1H, ArH), 7.61 (d, *J* = 8.2 Hz, 1H, ArH), 7.54 (t, *J* = 7.9 Hz, 1H, ArH), 7.38 (t, *J* = 8.9 Hz, 1H, ArH), 7.35 (t, *J* = 4.9 Hz, 2H, ArH), 7.29 (dd, *J* = 8.9, 1.4 Hz, 1H, ArH), 7.19 (dd, *J* = 5.6, 2.6 Hz, 1H, ArH), 4.68 (s, 2H, –NH–N<u>H</u>$_2$). ESI-MS m/z: 472.5 $[M+Na]^+$.

4.1.6.3.
*1-(3-Fluoro-4-((2-(hydrazinecarbonyl)pyridin-4-yl)oxy)phenyl)-3-(2-(trifluoro methyl)phenyl)urea (**11c**).* White solid; Yield: 76%; M.p. 202.3–205.0 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.92 (s, 1H, –N<u>H</u>–NH_2), 9.68 (s, 1H, NH), 8.49 (d, *J* = 5.5 Hz, 1H, ArH), 8.19 (s, 1H, NH), 7.90 (d, *J* = 7.8 Hz, 1H, ArH), 7.74 (dd, *J* = 13.6, 0.9 Hz, 1H, ArH), 7.69 (d, *J* = 7.8 Hz, 1H, ArH), 7.64 (d, *J* = 8.4 Hz, 1H, ArH), 7.38 (d, *J* = 8.5 Hz, 1H, ArH), 7.30–7.35 (m, 2H), 7.22 (dd, *J* = 8.6, 0.8 Hz, 1H), 7.16 (dd, *J* = 5.2, 1.8 Hz, 1H), 4.54 (s, 2H); ESI-MS m/z: 472.1 $[M+Na]^+$.

4.1.6.4.
*1-(3,4-Difluorophenyl)-3-(3-fluoro-4-((2-(hydrazinecarbonyl)pyridin-4-yl)oxy) phenyl)urea (**11d**).* White solid; Yield: 81%; M.p. 200.1–203.5 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.92 (s, 1H, –N<u>H</u>–NH_2), 9.24 (s, 1H, NH), 9.16 (s, 1H, NH), 8.49 (d, *J* = 5.6 Hz, 1H, ArH), 7.70 (dd, *J* = 13.3, 2.2 Hz, 1H, ArH), 7.62–7.68 (m, 1H, ArH), 7.30–7.38 (m, 3H, ArH), 7.26 (dd, *J* = 8.7, 0.9 Hz, 1H, ArH), 7.15 (dd, *J* = 5.6, 2.6 Hz, 2H, ArH), 4.54 (s, 2H, –NH–N<u>H</u>$_2$); ESI-MS m/z: 440.3 $[M+Na]^+$.

4.1.6.5.
*1-(3-Chloro-4-fluorophenyl)-3-(3-fluoro-4-((2-(hydrazinecarbonyl)pyridin-4-yl)oxy)phenyl)urea (**11e**).* White solid; Yield: 60%; M.p. 184.0–186.9 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.91 (s, 1H, –N<u>H</u>–NH_2), 9.68 (s, 1H, NH), 9.57 (s, 1H, NH), 8.49 (d, *J* = 2.0 Hz, 1H, ArH), 7.79–7.83 (m, 1H, ArH), 7.72 (d, *J* = 13.0 Hz, 1H, ArH), 7.34 (m, 4H, ArH), 7.28 (d, *J* = 8.6 Hz, 1H, ArH), 7.15 (d, *J* = 0.7 Hz, 1H, ArH), 4.54 (s, 2H, –NH–N<u>H</u>$_2$); ESI-MS m/z: 456.4 $[M+Na]^+$.

4.1.6.6.
*1-(2,4-Dichlorophenyl)-3-(3-fluoro-4-((2-(hydrazinecarbonyl)pyridin-4-yl)oxy) phenyl)urea (**11f**).* White solid; Yield: 67%; M.p. 222.9–224.0 °C; ^{1}H NMR (600 MHz, DMSO) δ 10.18 (s, 1H, –N<u>H</u>–NH_2), 9.79 (s, 1H, NH), 8.53 (d, *J* = 5.6 Hz, 1H, ArH), 8.51 (s, 1H, NH), 8.18 (d, *J* = 8.9 Hz, 1H, ArH), 7.75 (dd, *J* = 13.1, 2.4 Hz, 1H, ArH), 7.66 (d, *J* = 2.4 Hz, 1H, ArH), 7.42 (dd, *J* = 8.9, 2.4 Hz, 1H, ArH), 7.39 (d, *J* = 9.0 Hz, 1H, ArH), 7.37 (d, *J* = 2.5 Hz, 1H, ArH), 7.24 (dd, *J* = 8.8, 1.6 Hz, 1H, ArH), 7.20 (dd, *J* = 5.6, 2.6 Hz, 1H, ArH), 5.66 (s, 2H, –NH–N<u>H</u>$_2$); ESI-MS m/z: 450.3 $[M+H]^+$.

*4.1.7. General procedure for preparation of intermediates **12a–f***

A mixture of an appropriate hydrazine derivative (**11a–f**, 1 mmol) and DMF-DMA (5 mmol) in CH_2Cl_2 was heated under reflux for 4–6 h. The completion of the reaction was determined by TLC, at which point the solvent

was evaporated. Et_2O was added to the mixture, the precipitate was filtered off and washed to afford the solids **12a–f**.

4.1.7.1.
*(E)-N'-(4-(4-(3-(4-Chloro-3-(trifluoromethyl)phenyl)ureido)-2-fluorophenoxy)picolinoyl)-N,N-dimethylformohydrazonamide (**12a**).* Light yellow solid; Yield: 52%; M.p. 208.6–209.6 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.93 (s, 1H, –NH–N=CH), 9.33 (s, 1H, NH), 9.24 (s, 1H, NH), 8.49 (d, *J* = 5.6 Hz, 1H, ArH), 8.09 (d, *J* = 2.2 Hz, 1H, ArH), 8.03 (s, 1H, –NH–N=CH), 7.71 (dd, *J* = 13.1, 2.3 Hz, 1H, ArH), 7.66 (dd, *J* = 8.7, 2.1 Hz, 1H, ArH), 7.62 (d, *J* = 8.8 Hz, 1H, ArH), 7.36 (m, 2H, ArH), 7.28 (dd, *J* = 8.9, 1.7 Hz, 1H, ArH), 7.15 (dd, *J* = 5.6, 2.6 Hz, 1H, ArH), 2.80 (s, 6H, CH_3); ESI-MS m/z: 539.1 $[M+H]^+$.

4.1.7.2.
*(E)-N'-(4-(2-Fluoro-4-(3-(3-(trifluoromethyl)phenyl)ureido)phenoxy)picolinoyl)-N,N-dimethylformohydrazonamide (**12b**).* Light yellow solid; Yield: 66%; M.p. 201.0–203.2 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.92 (s, 1H, –NH–N=CH), 9.23 (s, 1H, NH), 9.21 (s, 1H, NH), 8.49 (s, 1H, –NH–N=CH), 8.02 (d, *J* = 10.9 Hz, 2H, ArH), 7.73 (dd, *J* = 12.2, 1.1 Hz, 1H, ArH), 7.55 (d, *J* = 30.9 Hz, 2H, ArH), 7.25–7.38 (m, 4H, ArH), 7.15 (s, 1H, ArH), 2.80 (s, 6H, CH_3); ESI-MS m/z: 505.1 $[M+H]^+$.

4.1.7.3.
*(E)-N'-(4-(2-Fluoro-4-(3-(2-(trifluoromethyl)phenyl)ureido)phenoxy)picolinoyl)-N,N-dimethylformohydrazonamide (**12c**).* Light yellow solid; Yield: 70%; M.p. 220.3–225.1 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.94 (s, 1H, –NH–N=CH), 9.68 (s, 1H, NH), 8.49 (d, *J* = 5.6 Hz, 1H, ArH), 8.19 (s, 1H, NH), 8.04 (s, 1H, –NH–N=CH), 7.90 (d, *J* = 8.3 Hz, 1H, ArH), 7.73 (dd, *J* = 13.2, 2.4 Hz, 1H, ArH), 7.69 (d, *J* = 7.9 Hz, 1H, ArH), 7.65 (t, *J* = 7.8 Hz, 1H, ArH), 7.34–7.39 (m, 2H, ArH), 7.30 (t, *J* = 7.6 Hz, 1H, ArH), 7.22 (dd, *J* = 8.8, 1.5 Hz, 1H, ArH), 7.15 (dd, *J* = 5.6, 2.6 Hz, 1H, ArH), 2.81 (s, 6H, CH_3); ESI-MS m/z: 505.2 $[M+H]^+$.

4.1.7.4.
*(E)-N'-(4-(4-(3-(3,4-Difluorophenyl)ureido)-2-fluorophenoxy)picolinoyl)-N,N-dimethylformohydrazonamide (**12d**).* Light yellow solid; Yield: 69%; M.p. 191.3–193.7 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.96 (s, 1H, –NH–N=CH), 9.13 (s, 1H, NH), 9.05 (s, 1H, NH), 8.50 (d, *J* = 5.6 Hz, 1H, ArH), 8.05 (s, 1H, –NH–N=CH), 7.70 (dd, *J* = 13.3, 2.4 Hz, 1H, ArH), 7.62–7.67 (m, 1H, ArH), 7.35 (m, 3H, ArH), 7.25 (dd, *J* = 8.9, 1.4 Hz, 1H, ArH), 7.15 (dt, *J* = 9.5, 4.8 Hz, 2H, ArH), 2.82 (s, 6H, CH_3); ESI-MS m/z: 473.1 $[M+H]^+$.

4.1.7.5.
*(E)-N'-(4-(4-(3-(3-Chloro-4-fluorophenyl)ureido)-2-fluorophenoxy)picolinoyl)-N,N-dimethylformohydrazonamide (**12e**).* Light yellow solid; Yield: 56%; M.p. 196.2–198.5 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.98 (s, 1H, –NH–N=CH), 9.17 (s, 1H, NH), 9.05 (s, 1H, NH), 8.50 (d, J = 5.7 Hz, 1H, ArH), 8.06 (s, 1H, –NH–N=CH), 7.79 (dd, *J* = 6.9, 1.7 Hz, 1H, ArH), 7.71 (dd, *J* = 13.5, 2.0 Hz, 1H, ArH), 7.31–7.36 (m, 4H, ArH), 7.25 (dd, *J* = 7.8, 1.5 Hz, 1H, ArH), 7.16 (dd, *J* = 5.4, 2.5 Hz, 1H, ArH), 2.83 (s, 6H, CH_3); ESI-MS m/z: 489.1 $[M+H]^+$.

4.1.7.6.
*(E)-N'-(4-(4-(3-(2,4-Dichlorophenyl)ureido)-2-fluorophenoxy)picolinoyl)-N,N-dimethylformohydrazonamide (**12f**).* Yellow solid; Yield: 77%; M.p. 172.7–175.0 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.95 (s, 1H, –N<u>H</u>–N=CH), 9.80 (s, 1H, NH), 8.52 (s, 1H, NH), 8.51 (br, 1H, ArH), 8.18 (d, *J* = 8.9 Hz, 1H, ArH), 8.05 (s, 1H, –NH–N=C<u>H</u>), 7.75 (dd, *J* = 13.2, 2.2 Hz, 1H, ArH), 7.66 (d, *J* = 2.3 Hz, 1H, ArH), 7.39–7.44 (m, 2H, ArH), 7.37 (d, *J* = 2.7 Hz,1H, ArH), 7.24 (dd, *J* = 8.3, 1.3 Hz, 1H, ArH), 7.17 (dd, *J* = 5.6, 2.6 Hz, 1H, ArH), 2.82 (s, 6H, CH_3); ESI-MS m/z: 505.2 $[M+H]^+$.

*4.1.8. General procedure for preparation of compounds **13a–y***

At room temperature, to a solution of an intermediate (**12a–f**, 0.3 mmol) in glacial acetic acid, an appropriate amine (0.9 mmol) was added slowly. The mixture was heated to 90 °C and stirred for 3 h. The solvent was evaporated under reduced pressure, and the residue was poured into water. The mixture was alkalized with aqueous NaOH to pH 8–9, at which time the precipitate was filtered off to yield the crude product. The solids were purified by silica gel column chromatography to generate the target compounds **13a–y**.

4.1.8.1.
*1-(4-Chloro-3-(trifluoromethyl)phenyl)-3-(3-fluoro-4-(2-(4-methyl-4H-1,2,4-triazol-3-yl)pyridin-4-yloxy)phenyl)urea (**13a**).* Flash column chromatography was performed using (dichloromethane:methanol, 100:1 to 25:1). HPLC purity: 98.7%; White solid; Yield: 51%; M.p. 179.8–181.3 °C; ^{1}H NMR (400 MHz, DMSO-d_6) 9.79 (s, 1H, NH), 9.68 (s, 1H, NH), 8.62 (d, *J* = 5.9 Hz, 2H, ArH), 8.11 (d, *J* = 2.1 Hz, 1H, ArH), 7.75 (dd, *J* = 13.2, 2.3 Hz, 1H, ArH), 7.68 (dd, *J* = 8.8, 2.1 Hz, 1H, ArH), 7.63 (d, *J* = 8.8 Hz, 1H, ArH), 7.55 (d, *J* = 2.4 Hz, 1H, ArH), 7.40 (t, *J* = 9.0 Hz, 1H, ArH), 7.29 (d, *J* = 8.8 Hz, 1H, ArH), 7.13 (dd, *J* = 5.7, 2.5 Hz, 1H, ArH), 4.00 (s, 3H, CH_3); ESI-MS m/z: 507.2 $[M+H]^+$; Anal. calcd. for $C_{22}H_{15}ClF_4N_6O_2$ (%): C, 52.13; H, 2.98; N, 16.58. Found (%): C, 51.37; H, 3.04; N, 16.72.

4.1.8.2.
*1-(3-Fluoro-4-(2-(4-methyl-4H-1,2,4-triazol-3-yl)pyridin-4-yloxy)phenyl)-3-(3-(trifluoromethyl)phenyl)urea (**13b**).* Flash column chromatography was performed using (dichloromethane:methanol, 150:1 to 25:1). HPLC purity: 99.2%; White solid; Yield: 37%; M.p. 231.0–232.3 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.20 (s, 2H, NH), 8.62 (s, 2H, NH), 8.03 (s, 1H, ArH), 7.76 (d, *J* = 10.9 Hz, 1H, ArH), 7.58 (m, 3H, ArH), 7.36 (m, 3H, ArH), 7.15 (s, 1H, ArH), 4.00 (s, 3H, CH_3); ^{13}C NMR (101 MHz, DMSO-d_6) δ 165.29, 154.02, 152.90, 151.79, 151.18, 149.76, 147.84, 140.78, 139.38, 134.27, 130.43, 130.01, 124.66, 124.45, 122.54, 118.83, 115.74, 114.82, 112.35, 109.12, 107.65, 34.17; ESI-MS m/z: 473.0 $[M+H]^+$; ESI-MS m/z: 473.0 $[M+H]^+$; Anal. calcd. for $C_{22}H_{16}F_4N_6O_2$ (%): C, 55.94; H, 3.41; N, 17.79. Found (%): C, 56.21; H, 3.25; N, 17.62.

4.1.8.3.
*1-(4-Chloro-3-(trifluoromethyl)phenyl)-3-(4-(2-(4-(2-(dimethylamino)ethyl)-4H-1,2,4-triazol-3-yl)pyridin-4-yloxy)-3-fluorophenyl)urea (**13c**).* Flash column

chromatography was performed using (dichloromethane:methanol, 70:1 to 15:1). HPLC purity: 98.7%; White solid; Yield: 33%; M.p. 178.6–180.5 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.97 (s, 1H, NH), 9.85 (s, 1H, NH), 8.64 (s, 1H, ArH), 8.60 (d, *J* = 5.2 Hz, 1H, ArH), 8.11 (s, 1H, ArH), 7.75 (d, *J* = 12.3 Hz, 1H, ArH), 7.65 (dd, *J* = 15.6, 8.1 Hz, 2H, ArH), 7.56 (s, 1H, ArH), 7.40 (t, *J* = 8.5 Hz, 1H, ArH), 7.28 (d, *J* = 8.0 Hz, 1H, ArH), 7.13 (dd, *J* = 5.6, 2.6 Hz, 1H, ArH), 4.61 (t, *J* = 5.9 Hz, 2H, –C$\underline{H}_2$–CH$_2$–N(CH$_3$)$_2$), 2.57 (t, *J* = 6.0 Hz, 2H, –CH$_2$–C$\underline{H}_2$–N(CH$_3$)$_2$), 2.13 (s, 6H, –CH$_2$–CH$_2$–N(C$\underline{H}_3$)$_2$); ^{13}C NMR (101 MHz, DMSO-d_6) δ 165.30, 154.04, 152.94, 151.72, 150.62, 150.06, 147.45, 139.67, 139.30, 134.35, 132.54, 127.24, 124.47, 123.79, 123.48, 121.91, 117.17, 115.60, 112.19, 109.28, 107.48, 59.24, 45.51, 43.84; ESI-MS m/z: 586.1 [M+Na]$^+$; Anal. calcd. for $C_{25}H_{22}ClF_4N_7O_2$ (%): C, 53.25; H, 3.93; N, 17.39. Found (%): C, 53.57; H, 4.15; N, 17.55.

4.1.8.4.
1-(4-(2-(4-(2-(Dimethylamino)ethyl)-4H-1,2,4-triazol-3-yl)pyridin-4-yloxy)-3-fluorophenyl)-3-(3-(trifluoromethyl)phenyl)urea ***(13d)***. Flash column chromatography was performed using (dichloromethane:methanol, 50:1 to 15:1). HPLC purity: 99.5%; White solid; Yield: 62%; M.p. 201.5–204.7 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.26 (s, 1H, NH), 9.24 (s, 1H, NH), 8.64 (s, 1H, ArH), 8.60 (d, *J* = 5.7 Hz, 1H, ArH), 8.03 (s, 1H, ArH), 7.76 (d, *J* = 13.2 Hz, 1H, ArH), 7.62 (d, *J* = 8.0 Hz, 1H, ArH), 7.49–7.56 (m, 2H, ArH), 7.40 (t, *J* = 8.9 Hz, 1H, ArH), 7.32 (m, 1H, ArH), 7.15 (dd, *J* = 5.7, 2.5 Hz, 1H, ArH), 4.63 (t, *J* = 6.0 Hz, 2H, –C$\underline{H}_2$–CH$_2$–N(CH$_3$)$_2$), 2.62 (t, *J* = 6.2 Hz, 2H, –CH$_2$–C$\underline{H}_2$–N(CH$_3$)$_2$), 2.17 (s, 6H, –CH$_2$–CH$_2$–N(C$\underline{H}_3$)$_2$); ^{13}C NMR (101 MHz, DMSO-d_6) δ 165.31, 154.02, 152.94, 151.72, 150.60, 150.05, 147.50, 140.82, 139.41, 134.27, 130.41, 139.99, 124.67, 124.45, 122.56, 118.82 115.76, 114.83, 112.29, 109.10, 107.65, 59.24, 45.53, 43.84; ESI-MS m/z: 530.2 [M+H]$^+$; Anal. calcd. for $C_{25}H_{23}F_4N_7O_2$ (%): C, 56.71; H, 4.38; N, 18.52. Found (%): C, 56.67; H, 4.65; N, 18.70.

4.1.8.5.
1-(4-(2-(4-(2-(Dimethylamino)ethyl)-4H-1,2,4-triazol-3-yl)pyridin-4-yloxy)-3-fluorophenyl)-3-(2-(trifluoromethyl)phenyl)urea ***(13e)***. Flash column chromatography was performed using (dichloromethane:methanol, 100:1 to 20:1). HPLC purity: 99.3%; White solid; Yield: 51%; M.p. 156.3–158.1 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.99 (s, 1H, NH), 8.64 (s, 1H, NH), 8.60 (s, 1H, ArH), 8.34 (s, 1H, ArH), 7.90 (s, 1H, ArH), 7.73 (m, 3H, ArH), 7.55 (s, 1H, ArH), 7.33 (m, 3H, ArH), 7.13 (s, 1H, ArH), 4.61 (br, 2H, –C$\underline{H}_2$–CH$_2$–N(CH$_3$)$_2$), 2.57 (br, 2H, –CH$_2$–C$\underline{H}_2$–N(CH$_3$)$_2$), 2.13 (s, 6H, –CH$_2$–CH$_2$–N(C$\underline{H}_3$)$_2$); ESI-MS m/z: 552.1 [M+Na]$^+$; Anal. calcd. for $C_{25}H_{23}F_4N_7O_2$ (%): C, 56.71; H, 4.38; N, 18.52. Found (%): C, 57.06; H, 4.17; N, 19.03.

4.1.8.6.
1-(3,4-Difluorophenyl)-3-(4-(2-(4-(2-(dimethylamino)ethyl)-4H-1,2,4-triazol-3-yl)pyridin-4-yloxy)-3-fluorophenyl)urea ***(13f)***. Flash column chromatography was performed using (dichloromethane:methanol, 50:1 to 15:1). HPLC purity: 99.7%; White solid; Yield: 46%; M.p. 165.2–167.8 °C; ^{1}H NMR (400 MHz,

DMSO-d_6) δ 9.13 (s, 1H, NH), 9.05 (s, 1H, NH), 8.59 (s, 1H, ArH), 8.56 (d, *J* = 5.8 Hz, 1H, ArH), 7.64 (m, 2H, ArH), 7.50 (d, *J* = 2.6 Hz, 1H, ArH), 7.35 (m, 2H, ArH), 7.25 (m, 1H, ArH), 7.11 (m, 2H, ArH), 4.58 (t, *J* = 6.4 Hz, 2H, –C<u>H</u>$_2$–CH$_2$–N(CH$_3$)$_2$), 2.55 (t, *J* = 6.4 Hz, 2H, –CH$_2$–C<u>H</u>$_2$–N(CH$_3$)$_2$), 2.10 (s, 6H, –CH$_2$–CH$_2$–N(C<u>H</u>$_3$)$_2$); ^{13}C NMR (101 MHz, DMSO-d_6) δ 165.33, 154.00, 153.06, 151.71, 150.60, 150.03, 147.48, 139.75, 137.36, 134.02, 124.38, 117.81, 115.53, 114.97, 112.20, 109.17, 107.82, 107.60, 107.37, 59.24, 45.53, 43.83; ESI-MS m/z: 498.1 $[M+H]^+$; Anal. calcd. for $C_{24}H_{22}F_3N_7O_2$ (%): C, 57.94; H, 4.46; N, 19.71. Found (%): C, 57.82; H, 4.57; N, 19.66.

4.1.8.7.
1-(3-Chloro-4-fluorophenyl)-3-(4-(2-(4-(2-(dimethylamino)ethyl)-4H-1,2,4-triazol-3-yl)pyridin-4-yloxy)-3-fluorophenyl)urea ***(13g)***. Flash column chromatography was performed using (dichloromethane:methanol, 60:1 to 15:1). HPLC purity: 99.2%; White solid; Yield: 61%; M.p. 206.1–209.6 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.23 (s, 1H, NH), 9.11 (s, 1H, NH), 8.63 (s, 1H, ArH), 8.59 (d, *J* = 5.6 Hz, 1H, ArH), 7.81 (d, *J* = 6.5 Hz, 1H, ArH), 7.74 (d, *J* = 12.5 Hz, 1H, ArH), 7.53 (d, *J* = 1.7 Hz, 1H, ArH), 7.37 (m, 3H, ArH), 7.29 (d, *J* = 8.6 Hz, 1H, ArH), 7.14 (dd, *J* = 5.3, 2.1 Hz, 1H, ArH), 4.61 (t, *J* = 5.9 Hz, 2H, –C<u>H</u>$_2$–CH$_2$–N(CH$_3$)$_2$), 2.57 (t, *J* = 6.1 Hz, 2H, –CH$_2$–C<u>H</u>$_2$–N(CH$_3$)$_2$), 2.12 (s, 6H, –CH$_2$–CH$_2$–N(C<u>H</u>$_3$)$_2$); ^{13}C NMR (101 MHz, DMSO-d_6) δ 165.34, 154.15, 154.00, 152.91, 151.73, 150.59, 149.97, 147.49, 139.52, 137.26, 134.15, 124.40, 120.26, 119.61, 119.25, 117.33, 115.65, 112.26, 109.12, 107.54, 58.97, 45.29, 43.61; ESI-MS m/z: 514.1 $[M+H]^+$; Anal. calcd. for $C_{24}H_{22}ClF_2N_7O_2$ (%): C, 56.09; H, 4.31; N, 19.08. Found (%): C, 56.36; H, 4.50; N, 19.37.

4.1.8.8.
1-(2,4-Dichlorophenyl)-3-(4-(2-(4-(2-(dimethylamino)ethyl)-4H-1,2,4-triazol-3-yl)pyridin-4-yloxy)-3-fluorophenyl)urea ***(13h)***. Flash column chromatography was performed using (dichloromethane:methanol, 50:1 to 20:1). HPLC purity: 98.9%; White solid; Yield: 52%; M.p. 183.6–185.8 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.06 (s, 1H, NH), 8.63 (s, 1H, NH), 8.60 (d, *J* = 5.4 Hz, 2H, ArH), 8.18 (d, *J* = 8.9 Hz, 1H, ArH), 7.76 (d, *J* = 11.5 Hz, 1H, ArH), 7.63 (s, 1H, ArH), 7.54 (s, 1H, ArH), 7.40 (m, 2H, ArH), 7.27 (d, *J* = 8.8 Hz, 1H, ArH), 7.13 (dd, *J* = 5.6, 2.5 Hz, 1H, ArH), 4.61 (t, *J* = 6.0 Hz, 2H, –C<u>H</u>$_2$–CH$_2$–N(CH$_3$)$_2$), 2.59 (t, *J* = 6.1 Hz, 2H, –CH$_2$–C<u>H</u>$_2$–N(CH$_3$)$_2$), 2.13 (s, 6H, –CH$_2$–CH$_2$–N(C<u>H</u>$_3$)$_2$); ^{13}C NMR (101 MHz, DMSO-d_6) δ 165.32, 154.06, 152.53, 151.75, 150.59, 149.95, 147.48, 139.39, 135.45, 134.25, 129.08, 128.08, 126.93, 124.56, 123.66, 123.03, 115.37, 112.24, 109.17, 107.21, 58.88, 45.22, 43.50; ESI-MS m/z: 552.0 $[M+Na]^+$; Anal. calcd. for $C_{24}H_{22}Cl_2FN_7O_2$ (%): C, 54.35; H, 4.18; N, 18.49. Found (%): C, 55.15; H, 4.17; N, 18.63.

4.1.8.9.
1-(4-Chloro-3-(trifluoromethyl)phenyl)-3-(4-(2-(4-(2-(diethylamino)ethyl)-4H-1,2,4-triazol-3-yl)pyridin-4-yloxy)-3-fluorophenyl)urea ***(13i)***. Flash column chromatography was performed using (dichloromethane:methanol, 100:1 to 20:1). HPLC purity: 99.0%; White solid; Yield: 41%; M.p. 202.2–205.6 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.40 (s, 1H, NH), 9.30 (s, 1H, NH), 8.62 (s, 1H,

ArH), 8.59 (d, *J* = 4.5 Hz, 1H, ArH), 8.12 (s, 1H, ArH), 7.70 (m, 3H, ArH), 7.52 (s, 1H, ArH), 7.36 (m, 2H, ArH), 7.14 (dd, *J* = 5.5 2.4 Hz, 1H, ArH), 4.56 (t, *J* = 5.9 Hz, 2H, –C$\underline{H}_2$–CH_2–N(CH_2CH_3)$_2$), 2.66 (t, *J* = 6.1 Hz, 2H, –CH_2–C$\underline{H}_2$–N(CH_2CH_3)$_2$), 2.39 (quartet, *J* = 6.3 Hz, 4H, –CH_2–CH_2–N(C$\underline{H}_2$$CH_3$)$_2$), 0.76 (t, *J* = 6.3 Hz, 6H, –CH_2–CH_2–N(CH_2C$\underline{H}_3$)$_2$); ^{13}C NMR (101 MHz, DMSO-*d*$_6$) δ 165.27, 154.00, 152.83, 151.67, 150.79, 150.21, 147.70, 139.57, 139.22, 134.40, 132.50, 127.19, 124.44, 123.79, 123.08, 121.91, 117.48, 115.90, 112.24, 109.08, 107.81, 53.08, 47.03, 44.45, 12.25; ESI-MS m/z: 592.0 $[M+H]^+$; Anal. calcd. for $C_{27}H_{26}ClF_4N_7O_2$ (%): C, 54.78; H, 4.43; N, 16.56. Found (%): C, 54.56; H, 4.15; N, 17.03.

4.1.8.10.
1-(4-(2-(4-(2-(Diethylamino)ethyl)-4H-1,2,4-triazol-3-yl)pyridin-4-yloxy)-3-fluorophenyl)-3-(3-(trifluoromethyl)phenyl)urea ***(13j)***. Flash column chromatography was performed using (dichloromethane:methanol, 100:1 to 25:1). HPLC purity: 99.3%; White solid; Yield: 36%; M.p. 181.3–183.5 °C; ^{1}H NMR (400 MHz, DMSO-*d*$_6$) δ 9.72 (s, 2H, NH), 8.62 (s, 1H, ArH), 8.58 (d, *J* = 5.6 Hz, 1H, ArH), 8.01 (s, 1H, ArH), 7.76 (dd, *J* = 13.0, 0.9 Hz, 1H, ArH), 7.62 (d, *J* = 8.2 Hz, 1H, ArH), 7.52 (m, 2H, ArH), 7.39 (t, *J* = 8.9 Hz, 1H, ArH), 7.30 (dd, *J* = 21.4, 7.9 Hz, 2H, ArH), 7.12 (dd, *J* = 5.1, 1.9 Hz, 1H, ArH), 4.55 (t, *J* = 5.8 Hz, 2H, –C$\underline{H}_2$–CH_2–N(CH_2CH_3)$_2$), 2.66 (t, *J* = 5.5 Hz, 2H, –CH_2–C$\underline{H}_2$–N(CH_2CH_3)$_2$), 2.39 (quartet, *J* = 6.1 Hz, 4H, –CH_2–CH_2–N(C$\underline{H}_2$$CH_3$)$_2$), 0.76 (t, *J* = 6.9 Hz, 6H, –CH_2–CH_2–N(CH_2C$\underline{H}_3$)$_2$); ^{13}C NMR (101 MHz, DMSO-*d*$_6$) δ 165.32, 154.05, 153.03, 151.68, 150.82, 150.22, 147.64, 140.89, 139.46, 134.24, 130.43, 130.04, 126.02, 124.44, 122.28, 118.71, 115.46, 114.59, 112.13, 109.29, 107.45, 53.12, 47.06, 44.46, 12.26; ESI-MS m/z: 580.7 $[M+Na]^+$; Anal. calcd. for $C_{27}H_{27}F_4N_7O_2$ (%): C, 58.16; H, 4.88; N, 17.59. Found (%): C, 58.37; H, 4.57; N, 18.03.

4.1.8.11.
1-(4-(2-(4-(2-(Diethylamino)ethyl)-4H-1,2,4-triazol-3-yl)pyridin-4-yloxy)-3-fluorophenyl)-3-(2-(trifluoromethyl)phenyl)urea ***(13k)***. Flash column chromatography was performed using (dichloromethane:methanol, 100:1 to 30:1). HPLC purity: 98.7%; White solid; Yield: 49%; M.p. 72.5–74.6 °C; ^{1}H NMR (400 MHz, DMSO-*d*$_6$) δ 9.94 (s, 1H, NH), 8.62 (s, 1H, NH), 8.58 (d, *J* = 5.6 Hz, 1H, ArH), 8.30 (s, 1H, ArH), 7.91 (d, *J* = 8.0 Hz, 1H, ArH), 7.75 (m, 3H, ArH), 7.54 (s, 1H, ArH), 7.38 (d, *J* = 9.1 Hz, 1H, ArH), 7.28 (m, 2H, ArH), 7.13 (dd, *J* = 5.6 2.5 Hz, 1H, ArH), 4.56 (t, *J* = 6.0 Hz, 2H, –C$\underline{H}_2$–CH_2–N(CH_2CH_3)$_2$), 2.68 (t, *J* = 5.9 Hz, 2H, –CH_2–C$\underline{H}_2$–N(CH_2CH_3)$_2$), 2.43 (quartet, *J* = 6.1 Hz, 4H, –CH_2–CH_2–N(C$\underline{H}_2$$CH_3$)$_2$), 0.79 (t, *J* = 6.3 Hz, 6H, –CH_2–CH_2–N(CH_2C$\underline{H}_3$)$_2$); ^{13}C NMR (101 MHz, DMSO-*d*$_6$) δ 165.30, 154.08, 152.99, 151.67, 150.80, 150.20, 147.68, 139.46, 136.41, 134.23, 133.41, 126.69, 126.45, 125.77, 124.61, 123.05, 120.98, 115.37, 112.19, 109.13, 107.28, 53.07, 47.04, 44.42, 12.22; ESI-MS m/z: 558.2 $[M+H]^+$; Anal. calcd. for $C_{27}H_{27}F_4N_7O_2$ (%): C, 58.16; H, 4.88; N, 17.59. Found (%): C, 58.22; H, 4.73; N, 17.31.

4.1.8.12.
1-(4-(2-(4-(2-(Diethylamino)ethyl)-4H-1,2,4-triazol-3-yl)pyridin-4-yloxy)-3-flu

*orophenyl)-3-(3,4-difluorophenyl)urea (**13l**)*. Flash column chromatography was performed using (dichloromethane:methanol, 60:1 to 20:1). HPLC purity: 99.1%; White solid; Yield: 32%; M.p. 211.3–212.6 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.16 (s, 1H, NH), 9.08 (s, 1H, NH), 8.66 (s, 1H, ArH), 8.60 (d, *J* = 5.8 Hz, 1H, ArH), 7.73 (dd, *J* = 13.3, 2.2 Hz, 1H, ArH), 7.67 (ddd, *J* = 13.2, 7.6, 2.5 Hz, 1H, ArH), 7.54 (s, 1H, ArH), 7.37 (m, 2H, ArH), 7.29 (d, *J* = 7.6 Hz, 1H, ArH), 7.16 (d, *J* = 8.0 Hz, 2H, ArH), 4.64 (t, *J* = 5.9 Hz, 2H, –C<u>H</u>$_2$–CH$_2$–N(CH$_2$CH$_3$)$_2$), 2.70 (t, *J* = 6.0 Hz, 2H, –CH$_2$–C<u>H</u>$_2$–N(CH$_2$CH$_3$)$_2$), 0.87 (t, *J* = 6.5 Hz, 6H, –CH$_2$–CH$_2$–N(CH$_2$C<u>H</u>$_3$)$_2$) (The protons of –CH$_2$–CH$_2$–N(C<u>H</u>$_2$CH$_3$)$_2$ overlapped with the peak of the solvent); ESI-MS m/z: 526.2 [M+H]+; Anal. calcd. for $C_{26}H_{26}F_3N_7O_2$ (%): C, 59.42; H, 4.99; N, 18.66. Found (%): C, 59.36; H, 4.93; N, 18.30.

4.1.8.13.
*1-(3-Chloro-4-fluorophenyl)-3-(4-(2-(4-(2-(diethylamino)ethyl)-4H-1,2,4-triazol-3-yl)pyridin-4-yloxy)-3-fluorophenyl)urea (**13m**)*. Flash column chromatography was performed using (dichloromethane:methanol, 80:1 to 25:1). HPLC purity: 99.2%; White solid; Yield: 55%; M.p. 146.5–148.7 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.45 (s, 1H, NH), 9.34 (s, 1H, NH), 8.62 (s, 1H, ArH), 8.58 (d, *J* = 5.7 Hz, 1H, ArH), 7.79–7.82 (dd, *J* = 7.6, 2.0 Hz, 1H, ArH), 7.74 (dd, *J* = 13.2, 2.3 Hz, 1H, ArH), 7.52 (d, *J* = 2.4 Hz, 1H, ArH), 7.39 (t, *J* = 9.0 Hz, 1H, ArH), 7.35–7.36 (m, 1H, ArH), 7.33 (m, 1H, ArH), 7.27 (dd, *J* = 8.8, 1.3 Hz, 1H, ArH), 7.13 (dd, *J* = 5.7, 2.5 Hz, 1H, ArH), 4.55 (t, *J* = 6.2 Hz, 2H, –C<u>H</u>$_2$–CH$_2$–N(CH$_2$CH$_3$)$_2$), 2.66 (t, *J* = 6.0 Hz, 2H, –CH$_2$–C<u>H</u>$_2$–N(CH$_2$CH$_3$)$_2$), 2.39 (quartet, *J* = 6.9 Hz, 4H, –CH$_2$–CH$_2$–N(C<u>H</u>$_2$CH$_3$)$_2$), 0.76 (t, *J* = 7.0 Hz, 6H, –CH$_2$–CH$_2$–N(CH$_2$C<u>H</u>$_3$)$_2$); ^{13}C NMR (101 MHz, DMSO-d_6) δ 165.31, 154.04, 152.97, 152.95, 151.67, 150.81, 150.22, 147.65, 139.48, 137.26, 134.22, 124.43, 120.14, 119.65, 119.13, 117.39, 115.52, 112.18, 109.21, 107.42, 53.11, 47.06, 44.45, 12.25; ESI-MS m/z: 542.2 [M+H]$^+$; Anal. calcd. for $C_{26}H_{26}ClF_2N_7O_2$ (%): C, 57.62; H, 4.84; N, 18.09. Found (%): C, 58.01; H, 4.67; N, 18.23.

4.1.8.14.
*1-(2,4-Dichlorophenyl)-3-(4-(2-(4-(2-(diethylamino)ethyl)-4H-1,2,4-triazol-3-yl)pyridin-4-yloxy)-3-fluorophenyl)urea (**13n**)*. Flash column chromatography was performed using (dichloromethane:methanol, 100:1 to 30:1). HPLC purity: 99.5%; White solid; Yield: 37%; M.p. 139.1–142.1 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.87 (s, 1H, NH), 8.62 (s, 1H, NH), 8.59 (d, *J* = 5.7 Hz, 1H, ArH), 8.53 (s, 1H, ArH), 8.18 (d, *J* = 9.0 Hz, 1H, ArH), 7.76 (d, *J* = 13.3 Hz, 1H, ArH), 7.64 (d, *J* = 1.9 Hz, 1H, ArH), 7.53 (d, *J* = 1.4 Hz, 1H, ArH), 7.40 (m, 2H, ArH), 7.26 (d, *J* = 9.1 Hz, 1H, ArH), 7.14 (dd, *J* = 5.6, 2.4 Hz, 1H, ArH), 4.56 (t, *J* = 6.0 Hz, 2H, –C<u>H</u>$_2$–CH$_2$–N(CH$_2$CH$_3$)$_2$), 2.68 (t, *J* = 6.1 Hz, 2H, –CH$_2$–C<u>H</u>$_2$–N(CH$_2$CH$_3$)$_2$), 2.42 (quartet, *J* = 5.9 Hz, 4H, –CH$_2$–CH$_2$–N(C<u>H</u>$_2$CH$_3$)$_2$), 0.78 (t, *J* = 6.7 Hz, 6H, –CH$_2$–CH$_2$–N(CH$_2$C<u>H</u>$_3$)$_2$); ^{13}C NMR (101 MHz, DMSO-d_6) δ 165.30, 154.07, 152.45, 151.70, 150.76, 150.12, 147.67, 139.22, 135.38, 134.35, 129.10, 128.16, 127.00, 124.60, 123.54, 122.93, 115.48, 112.27, 109.09, 107.36, 52.87, 47.10, 29.48, 12.05; ESI-MS m/z: 558.0 [M+H]$^+$; Anal. calcd. for $C_{26}H_{26}Cl_2FN_7O_2$ (%): C, 55.92; H, 4.69; N, 17.56. Found (%): C, 56.11; H, 4.53; N, 17.53.

4.1.8.15.

*1-(4-Chloro-3-(trifluoromethyl)phenyl)-3-(4-(2-(4-(3-(dimethylamino)propyl)-4H-1,2,4-triazol-3-yl)pyridin-4-yloxy)-3-fluorophenyl)urea (**13o**).* Flash column chromatography was performed using (dichloromethane:methanol, 50:1 to 8:1). HPLC purity: 99.1%; White solid; Yield: 59%; M.p. 182.6–185.3 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.63 (s, 1H, NH), 9.53 (s, 1H, NH), 8.66 (s, 1H, ArH), 8.60 (d, *J* = 5.7 Hz, 1H, ArH), 8.12 (d, *J* = 2.2 Hz, 1H, ArH), 7.75 (dd, *J* = 13.1, 2.2 Hz, 1H, ArH), 7.68 (dd, *J* = 8.8, 2.1 Hz, 1H, ArH), 7.63 (d, *J* = 8.8 Hz, 1H, ArH), 7.56 (d, *J* = 2.4 Hz, 1H, ArH), 7.40 (t, *J* = 8.9 Hz, 1H, ArH), 7.30 (d, *J* = 8.7 Hz, 1H, ArH), 7.13 (dd, *J* = 5.7, 2.5 Hz, 1H, ArH), 4.52 (t, *J* = 7.2 Hz, 2H, –C<u>H</u>$_2$–CH$_2$–CH$_2$–N(CH$_3$)$_2$), 2.20 (t, *J* = 6.7 Hz, 2H, –CH$_2$–CH$_2$–C<u>H</u>$_2$–N(CH$_3$)$_2$), 2.11 (s, 6H, –CH$_2$–CH$_2$–CH$_2$–N(C<u>H</u>$_3$)$_2$), 1.88 (quintet, *J* = 7.0 Hz, 2H, –CH$_2$–C<u>H</u>$_2$–CH$_2$–N(CH$_3$)$_2$); ESI-MS m/z: 578.1 $[M+H]^+$; Anal. calcd. for $C_{26}H_{24}ClF_4N_7O_2$ (%): C, 54.03; H, 4.19; N, 16.96. Found (%): C, 54.32; H, 4.22; N, 16.67.

4.1.8.16.

*1-(4-(2-(4-(3-(Dimethylamino)propyl)-4H-1,2,4-triazol-3-yl)pyridin-4-yloxy)-3-fluorophenyl)-3-(3-(trifluoromethyl)phenyl)urea (**13p**).* Flash column chromatography was performed using (dichloromethane:methanol, 50:1 to 10:1). HPLC purity: 98.8%; White solid; Yield: 29%; M.p. 176.0–178.3 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.92 (s, 2H, NH), 8.65 (s, 1H, ArH), 8.59 (d, *J* = 5.7 Hz, 1H, ArH), 8.04 (s, 1H, ArH), 7.77 (d, *J* = 13.2 Hz, 1H, ArH), 7.65 (d, *J* = 8.1 Hz, 1H, ArH), 7.55 (d, *J* = 2.4 Hz, 1H, ArH), 7.51 (t, *J* = 7.9 Hz, 1H, ArH), 7.35 (m, 3H, ArH), 7.12 (dd, *J* = 5.7, 2.5 Hz, 1H, ArH), 4.50 (t, *J* = 7.2 Hz, 2H, –C<u>H</u>$_2$–CH$_2$–CH$_2$–N(CH$_3$)$_2$), 2.16 (t, *J* = 6.7 Hz, 2H, –CH$_2$–CH$_2$–C<u>H</u>$_2$–N(CH$_3$)$_2$), 2.08 (s, 6H, –CH$_2$–CH$_2$–CH$_2$–N(C<u>H</u>$_3$)$_2$), 1.85 (quintet, *J* = 6.9 Hz, 2H, –CH$_2$–C<u>H</u>$_2$–CH$_2$–N(CH$_3$)$_2$); ESI-MS m/z: 544.2 $[M+H]^+$; Anal. calcd. for $C_{26}H_{25}F_4N_7O_2$ (%): C, 57.46; H, 4.64; N, 18.04. Found (%): C, 57.62; H, 4.53; N, 18.38.

4.1.8.17.

*1-(4-(2-(4-(3-(Dimethylamino)propyl)-4H-1,2,4-triazol-3-yl)pyridin-4-yloxy)-3-fluorophenyl)-3-(2-(trifluoromethyl)phenyl)urea (**13q**).* Flash column chromatography was performed using (dichloromethane:methanol, 30:1 to 7:1). HPLC purity: 99.7%; White solid; Yield: 50%; M.p. 152.6–159.3 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.60 (s, 1H, ArH), 8.56 (d, *J* = 5.8 Hz, 1H, ArH), 7.87 (d, *J* = 8.2 Hz, 1H, ArH), 7.75 (dd, *J* = 13.4, 2.3 Hz, 1H, ArH), 7.61 (dd, *J* = 17.7, 8.2 Hz, 2H, ArH), 7.52 (d, *J* = 2.5 Hz, 1H, ArH), 7.34 (t, *J* = 9.0 Hz, 1H, ArH), 7.24 (m, 2H, ArH), 7.08 (dd, *J* = 5.7, 2.5 Hz, 1H, ArH), 4.47 (t, *J* = 7.2 Hz, 2H, –C<u>H</u>$_2$–CH$_2$–CH$_2$–N(CH$_3$)$_2$), 2.12 (t, *J* = 6.7 Hz, 2H, –CH$_2$–CH$_2$–C<u>H</u>$_2$–N(CH$_3$)$_2$), 2.04 (s, 6H, –CH$_2$–CH$_2$–CH$_2$–N(C<u>H</u>$_3$)$_2$), 1.82 (quintet, *J* = 7.0 Hz, 2H, –CH$_2$–C<u>H</u>$_2$–CH$_2$–N(CH$_3$)$_2$) (Protons of NH was disappeared); ^{13}C NMR (101 MHz, DMSO-d_6) δ 165.30, 154.07, 153.08, 151.78, 150.73, 149.98, 147.35, 139.55, 136.48, 134.18, 133.41, 126.85, 126.47, 124.71, 124.56, 123.05, 121.16, 115.35, 112.24, 109.16, 107.27, 56.20, 45.42, 44.53, 28.64; ESI-MS m/z: 566.1 $[M+Na]^+$; Anal. calcd. for $C_{26}H_{25}F_4N_7O_2$ (%): C, 57.46; H, 4.64; N, 18.04. Found (%): C, 57.57; H, 4.33; N, 17.96.

4.1.8.18.
*1-(3,4-Difluorophenyl)-3-(4-(2-(4-(3-(dimethylamino)propyl)-4H-1,2,4-triazol-3-yl)pyridin-4-yloxy)-3-fluorophenyl)urea (**13r**).* Flash column chromatography was performed using (dichloromethane:methanol, 50:1 to 10:1). HPLC purity: 99.1%; White solid; Yield: 36%; M.p. 177.1–180.2 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.36 (s, 1H, NH), 9.28 (s, 1H, NH), 8.65 (s, 1H, ArH), 8.60 (d, *J* = 5.7 Hz, 1H, ArH), 7.74 (dd, *J* = 13.2, 1.7 Hz, 1H, ArH), 7.68 (ddd, *J* = 13.2, 7.5, 2.2 Hz, 1H, ArH), 7.55 (d, *J* = 2.1 Hz, 1H, ArH), 7.38 (dd, *J* = 18.0, 9.1 Hz, 2H, ArH), 7.30 (t, *J* = 8.2 Hz, 1H, ArH), 7.17 (d, *J* = 9.0 Hz, 1H, ArH), 7.13 (dd, *J* = 5.6, 2.4 Hz, 1H, ArH), 4.51 (t, *J* = 7.2 Hz, 2H, –C<u>H</u>$_2$–CH$_2$–CH$_2$–N(CH$_3$)$_2$), 2.16 (t, *J* = 6.6 Hz, 2H, –CH$_2$–CH$_2$–C<u>H</u>$_2$–N(CH$_3$)$_2$), 2.09 (s, 6H, –CH$_2$–CH$_2$–CH$_2$–N(C<u>H</u>$_3$)$_2$), 1.85 (quintet, *J* = 7.2 Hz, 2H, –CH$_2$–C<u>H</u>$_2$–CH$_2$–N(CH$_3$)$_2$); ^{13}C NMR (101 MHz, DMSO-d_6) δ 165.29, 154.00, 152.87, 151.78, 150.72, 149.97, 147.35, 139.45, 137.00, 134.19, 124.45, 117.87, 115.66, 115.13, 112.28, 109.10, 108.01, 107.80, 107.55, 99.99, 56.14, 45.33, 44.50, 28.54; ESI-MS m/z: 512.1; [M+H]$^+$; Anal. calcd. for $C_{25}H_{24}F_3N_7O_2$ (%): C, 58.70; H, 4.73; N, 19.17. Found (%): C, 59.03; H, 4.59; N, 19.56.

4.1.8.19.
*1-(3-Chloro-4-fluorophenyl)-3-(4-(2-(4-(3-(dimethylamino)propyl)-4H-1,2,4-triazol-3-yl)pyridin-4-yloxy)-3-fluorophenyl)urea (**13s**).* Flash column chromatography was performed using (dichloromethane:methanol, 60:1 to 8:1). HPLC purity: 99.5%; White solid; Yield: 52%; M.p. 173.5–176.3 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.32 (s, 1H, NH), 9.21 (s, 1H, NH), 8.66 (s, 1H, ArH), 8.60 (d, *J* = 5.7 Hz, 1H, ArH), 7.81 (d, *J* = 7.2 Hz, 1H, ArH), 7.74 (dd, *J* = 13.2, 2.0 Hz, 1H, ArH), 7.55 (d, *J* = 2.4 Hz, 1H, ArH), 7.34 (m, 4H, ArH), 7.13 (dd, *J* = 5.7, 2.5 Hz, 1H, ArH), 4.51 (t, *J* = 7.3 Hz, 2H, –C<u>H</u>$_2$–CH$_2$–CH$_2$–N(CH$_3$)$_2$), 2.16 (t, *J* = 6.7 Hz, 2H, –CH$_2$–CH$_2$–C<u>H</u>$_2$–N(CH$_3$)$_2$), 2.09 (s, 6H, –CH$_2$–CH$_2$–CH$_2$–N(C<u>H</u>$_3$)$_2$), 1.85 (quintet, *J* = 6.8 Hz, 2H, –CH$_2$–C<u>H</u>$_2$–CH$_2$–N(CH$_3$)$_2$); ESI-MS m/z: 528.3; [M+H]$^+$; Anal. calcd. for $C_{25}H_{24}ClF_2N_7O_2$ (%): C, 56.87; H, 4.58; N, 18.57. Found (%): C, 58.63; H, 4.70; N, 18.30.

4.1.8.20.
*1-(2,4-Dichlorophenyl)-3-(4-(2-(4-(3-(dimethylamino)propyl)-4H-1,2,4-triazol-3-yl)pyridin-4-yloxy)-3-fluorophenyl)urea (**13t**).* Flash column chromatography was performed using (dichloromethane:methanol, 30:1 to 7:1). HPLC purity: 99.2%; White solid; Yield: 43%; M.p. 181.9–184.2 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.79 (s, 1H, NH), 8.65 (s, 1H, NH), 8.61 (d, *J* = 5.7 Hz, 1H, ArH), 8.50 (s, 1H, ArH), 8.18 (d, *J* = 9.0 Hz, 1H, ArH), 7.76 (dd, *J* = 13.2, 2.4 Hz, 1H, ArH), 7.64 (d, *J* = 2.4 Hz, 1H, ArH), 7.55 (d, *J* = 2.5 Hz, 1H, ArH), 7.41 (m, 2H, ArH), 7.26 (d, *J* = 8.9 Hz, 1H, ArH), 7.14 (dd, *J* = 5.7, 2.6 Hz, 1H, ArH), 4.52 (t, *J* = 7.2 Hz, 2H, –C<u>H</u>$_2$–CH$_2$–CH$_2$–N(CH$_3$)$_2$), 2.28 (t, *J* = 6.7 Hz, 2H, –CH$_2$–CH$_2$–C<u>H</u>$_2$–N(CH$_3$)$_2$), 2.17 (s, 6H, –CH$_2$–CH$_2$–CH$_2$–N(C<u>H</u>$_3$)$_2$), 1.89 (quintet, *J* = 6.9 Hz, 2H, –CH$_2$–C<u>H</u>$_2$–CH$_2$–N(CH$_3$)$_2$); ^{13}C NMR (101 MHz, DMSO-d_6) δ 165.27, 154.08, 152.48, 151.79, 150.72, 149.98, 147.36, 139.24, 135.41, 134.35, 129.10, 128.15, 127.03, 124.60, 123.63, 123.03, 115.48, 112.28, 109.12, 107.39, 56.20, 45.42, 44.54, 28.64; ESI-MS m/z: 544.0

$[M+H]^+$; Anal. calcd. for $C_{25}H_{24}Cl_2FN_7O_2$ (%): C, 55.15; H, 4.44; N, 18.01. Found (%): C, 55.07; H, 4.52; N, 18.34.

4.1.8.21.
*1-(4-Chloro-3-(trifluoromethyl)phenyl)-3-(3-fluoro-4-((2-(4-(3-morpholinopropyl)-4H-1,2,4-triazol-3-yl)pyridin-4-yl)oxy)phenyl)urea (**13u**).* Flash column chromatography was performed using (dichloromethane:methanol, 60:1 to 20:1). HPLC purity: 99.3%; White solid; Yield: 38%; M.p. 150.5–152.9 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.71 (s, 1H, NH), 9.60 (s, 1H, NH), 8.66 (s, 1H, ArH), 8.58 (d, *J* = 5.7 Hz, 1H, ArH), 8.11 (d, *J* = 1.4 Hz, 1H, ArH), 7.74 (dd, *J* = 13.1, 1.5 Hz, 1H, ArH), 7.68 (dd, *J* = 8.6, 0.9 Hz, 1H, ArH), 7.62 (d, *J* = 8.7 Hz, 1H, ArH), 7.54 (d, *J* = 2.0 Hz, 1H, ArH), 7.39 (t, *J* = 8.9 Hz, 1H, ArH), 7.30 (d, *J* = 8.7 Hz, 1H, ArH), 7.12 (dd, *J* = 5.5, 2.3 Hz, 1H, ArH), 4.54 (t, *J* = 6.9 Hz, 2H, –C<u>H</u>$_2$–CH$_2$–CH$_2$–(C_4H_8NO)), 3.50 (br, 4H, morpholinyl), 2.19–2.25 (m, 6H, –CH$_2$–CH$_2$–C<u>H</u>$_2$–(C_4H_8NO), morpholinyl), 1.89 (quintet, *J* = 7.1 Hz, 2H, –CH$_2$–C<u>H</u>$_2$–CH$_2$–(C_4H_8NO)); ESI-MS m/z: 642.1 $[M+Na]^+$; Anal. calcd. for $C_{28}H_{26}ClF_4N_7O_3$ (%): C, 54.24; H, 4.23; N, 15.81. Found (%): C, 54.30; H, 4.16; N, 16.06.

4.1.8.22.
*1-(3-Fluoro-4-((2-(4-(3-morpholinopropyl)-4H-1,2,4-triazol-3-yl)pyridin-4-yl)oxy)phenyl)-3-(2-(trifluoromethyl)phenyl)urea (**13v**).* Flash column chromatography was performed using (dichloromethane:methanol, 100:1 to 20:1). HPLC purity: 98.9%; White solid; Yield: 51%; M.p. 138.1–140.2 °C; ^{1}H NMR (300 MHz, DMSO-d_6) δ 9.77 (s, 1H, NH), 8.63 (s, 1H, NH), 8.54 (d, *J* = 5.8 Hz, 1H, ArH), 8.21 (s, 1H, ArH), 7.88 (d, *J* = 8.2 Hz, 1H, ArH), 7.72 (dd, *J* = 13.2, 2.2 Hz, 1H, ArH), 7.64 (dd, *J* = 16.3, 8.0 Hz, 2H, ArH), 7.51 (d, *J* = 2.4 Hz, 1H, ArH), 7.36 (t, *J* = 9.0 Hz, 1H, ArH), 7.28 (t, *J* = 7.7 Hz, 1H, ArH), 7.22 (dd, *J* = 8.9, 1.4 Hz, 1H, ArH), 7.10 (dd, *J* = 5.6, 2.5 Hz, 1H, ArH), 4.51 (t, *J* = 7.0 Hz, 2H, –C<u>H</u>$_2$–CH$_2$–CH$_2$–(C_4H_8NO)), 3.47 (br, 4H, morpholinyl), 2.22 (m, 6H, –CH$_2$–CH$_2$–C<u>H</u>$_2$–(C_4H_8NO), morpholinyl), 1.87 (quintet, *J* = 6.8 Hz, 2H, –CH$_2$–C<u>H</u>$_2$–CH$_2$–(C_4H_8NO)); ^{13}C NMR (101 MHz, DMSO-d_6) δ 165.30, 154.08, 153.02, 151.73, 150.79, 150.04, 147.37, 139.49, 136.41, 134.20, 133.42, 126.76, 126.45, 125.76, 124.63, 123.05, 121.04, 115.36, 112.26, 109.18, 107.26, 66.55, 55.14, 53.51, 44.38, 27.30; ESI-MS m/z: 608.1 $[M+Na]^+$; Anal. calcd. for $C_{28}H_{27}F_4N_7O_3$ (%): C, 57.43; H, 4.65; N, 16.74. Found (%): C, 57.50; H, 4.42; N, 16.67.

4.1.8.23.
*1-(3-Chloro-4-fluorophenyl)-3-(3-fluoro-4-((2-(4-(3-morpholinopropyl)-4H-1,2,4-triazol-3-yl)pyridin-4-yl)oxy)phenyl)urea (**13w**).* Flash column chromatography was performed using (dichloromethane:methanol, 60:1 to 20:1). HPLC purity: 99.2%; White solid; Yield: 47%; M.p. 162.6–165.2 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.43 (s, 1H, NH), 9.32 (s, 1H, NH), 8.65 (s, 1H, ArH), 8.56 (d, *J* = 5.7 Hz, 1H, ArH), 7.79 (dd, *J* = 6.1, 1.3 Hz, 1H, ArH), 7.72 (dd, *J* = 13.1, 1.9 Hz, 1H, ArH), 7.52 (d, *J* = 1.7 Hz, 1H, ArH), 7.30–7.40 (m, 3H, ArH), 7.24 (dd, *J* = 8.5, 0.7 Hz, 1H, ArH), 7.12 (dd, *J* = 5.5, 2.4 Hz, 1H, ArH), 4.52 (t, *J* = 6.9 Hz, 2H, –C<u>H</u>$_2$–CH$_2$–CH$_2$–(C_4H_8NO)), 3.49 (br, 4H, morpholinyl), 2.23 (m, 6H, –CH$_2$–CH$_2$–C<u>H</u>$_2$–(C_4H_8NO), morpholinyl), 1.87

(quintet, *J* = 7.0 Hz, 2H, –CH_2–C<u>H</u>$_2$–CH_2–(C_4H_8NO)); ^{13}C NMR (101 MHz, DMSO-d_6) 165.30, 154.14, 154.03, 152.94, 151.74, 150.78, 150.02, 147.37, 139.49, 137.24, 134.14, 124.49, 120.08, 119.65, 119.10, 117.42, 115.50, 112.25, 109.17, 107.38, 66.50, 55.09, 53.47, 44.36, 27.25; ESI-MS m/z: 569.8 $[M+H]^+$; Anal. calcd. for $C_{28}H_{27}F_4N_7O_3$ (%): C, 56.89; H, 4.60; N, 17.20. Found (%): C, 56.95; H, 4.55; N, 17.32.

4.1.8.24.
*1-(2,4-dichlorophenyl)-3-(3-fluoro-4-((2-(4-(3-morpholinopropyl)-4H-1,2,4-triazol-3-yl)pyridin-4-yl)oxy)phenyl)urea (**13x**)*. Flash column chromatography was performed using (dichloromethane:methanol, 100:1 to 30:1). HPLC purity: 99.4%; White solid; Yield: 66%; M.p. 156.8–160.2 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.97 (s, 1H, NH), 8.65 (s, 1H, NH), 8.55–8.58 (m, 2H, ArH), 8.16 (d, *J* = 9.0 Hz, 1H, ArH), 7.75 (dd, *J* = 13.2, 2.3 Hz, 1H, ArH), 7.63 (d, *J* = 2.4 Hz, 1H, ArH), 7.52 (d, *J* = 2.3 Hz, 1H, ArH), 7.39 (ddd, *J* = 8.9, 5.7, 3.1 Hz, 2H, ArH), 7.24 (dd, *J* = 8.8, 1.0 Hz, 1H, ArH), 7.13 (dd, *J* = 5.7, 2.5 Hz, 1H, ArH), 4.53 (t, *J* = 7.0 Hz, 2H, –C<u>H</u>$_2$–CH_2–CH_2–(C_4H_8NO)), 3.49 (br, 4H, morpholinyl), 2.23 (m, 6H, –CH_2–CH_2–C<u>H</u>$_2$–(C_4H_8NO), morpholinyl), 1.87 (br, 2H, –CH_2–C<u>H</u>$_2$–CH_2–(C_4H_8NO)); ^{13}C NMR (101 MHz, DMSO-d_6) δ 165.29, 154.08, 152.47, 151.78, 150.73, 149.97, 147.35, 139.20, 135.39, 134.33, 129.11, 128.15, 127.01, 124.61, 123.58, 122.97, 115.49, 112.32, 109.12, 107.36, 66.14, 53.20, 44.25; ESI-MS m/z: 586.0 $[M+H]^+$; Anal. calcd. for $C_{28}H_{27}F_4N_7O_3$ (%): C, 55.30; H, 4.47; N, 16.72. Found (%): C, 55.22; H, 4.68; N, 16.65.

4.1.8.25.
*1-(4-Chloro-3-(trifluoromethyl)phenyl)-3-(4-(2-(4-cyclopropyl-4H-1,2,4-triazol-3-yl)pyridin-4-yloxy)-3-fluorophenyl)urea (**13y**)*. Flash column chromatography was performed using (dichloromethane:methanol, 100:1 to 25:1). HPLC purity: 98.5%; White solid; Yield: 58%; M.p. 171.1–173.5 °C; ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.47 (s, 1H, NH), 10.33 (s, 1H, NH), 8.66 (s, 1H, ArH), 8.63 (d, *J* = 5.6 Hz, 1H, ArH), 8.11 (s, 1H, ArH), 7.76 (dd, *J* = 13.0, 1.9 Hz, 1H, ArH), 7.65 (m, 2H, ArH), 7.50 (d, *J* = 1.9 Hz, 1H, ArH), 7.40 (t, *J* = 9.0 Hz, 1H, ArH), 7.26 (d, *J* = 8.0 Hz, 1H, ArH), 7.12 (dd, *J* = 5.5, 2.2 Hz, 1H, ArH), 3.51 (br, 1H, cyclopropyl), 0.96 (m, 2H, cyclopropyl), 0.84 (m, 2H, cyclopropyl); ESI-MS m/z: 533.3 $[M+H]^+$; Anal. calcd. for $C_{24}H_{17}ClF_4N_6O_2$ (%): C, 54.09; H, 3.22; N, 15.77. Found (%): C, 53.89; H, 3.37; N, 15.65.

4.2. Pharmacology

4.2.1. MTT assay

MTT assay was carried out on H460, HT-29, MDA-MB-231, and WI-38 cells to evaluate the cytotoxicity of the newly synthesized compounds. The cancer cell lines were cultured in minimum essential medium (MEM) supplement with 10% foetal bovine serum (FBS).

Approximate 4×10^3 cells, suspended in MEM medium, were plated in a 96-well plate and incubated in 5% CO_2 at 37 °C for 24 h. The tested compounds were added to the culture medium and the cell cultures were incubated for another 72 h. Fresh MTT was added to each well at a terminal

concentration of 5 μg/mL, and incubated with cells at 37 °C for 4 h. The formazan crystals in each well were dissolved in 100 μL DMSO, and the absorbency at 492 nm (absorbance of MTT formazan) and 630 nm (reference wavelength) was measured with an ELISA reader. All of the compounds were tested three times in each cell line. The results, expressed as inhibitory concentration 50 % (IC_{50}), are averages of at least three determinations and calculated using the Bacus Laboratories Incorporated Slide Scanner (Bliss) software.

4.2.2. Enzymatic activity assay

The tested compounds were diluted to 50-fold of the final highest desired concentration by 100 % DMSO. Each well of a 96-well plate received 5 μL of compound and 95 μL of 100 % DMSO. Two wells received only 100 μL of 100% DMSO and served as negative controls (no compound and no enzyme). This plate was labelled the "source plate." Subsequently, 10 μL of compound from the "source plate" was transferred to a new 96-well plate named the "intermediate plate," and 90 μL of kinase buffer was added. Then, 5 μL from each well of the "intermediate plate" was transferred in duplicate to a 384-well plate. To each well, fresh enzyme and peptide solutions were added successively. The mixture was incubated at 28 °C for 1 h, then 25 μL of stop buffer was added. The biological data was collected from Caliper program.

4.2.3. Apoptosis analysis

HT-29 cells (5×10^5 cells/mL) were seeded in six-well plates and treated with compound **13i** at different concentrations for 48 h. The cells were then harvested by trypsinization and washed twice with cold PBS. After centrifugation and removal of the supernatants, cells were resuspended in 400 μL of 1 × binding buffer which was then added to 5 μL of annexinV-FITC and incubated at room temperature for 15 min. After adding 10 μL of PI the cells were incubated at room temperature for another 15 min in the dark. The stained cells were analyzed by a flow cytometer (BD Accuri C6).

Conflict of interest

The authors have declared no conflict of interest.

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Legends

Figure 1. Design strategy of the target compounds.
Figure 2. Effect of compound **13i** on cell apoptosis in HT-29 cells. FL1-H: AnnexinV-FITC; FL2-H: PI.

Scheme 1. Reagents and conditions: (a) triphosgene, toluene, reflux, 7–10 h; (b) (i) $SOCl_2$, NaBr, chlorobenzene, 50 °C, 30 min, 85 °C, 20 h; (ii) MeOH, toluene, 0–15 °C, 1.5 h; (c) 2-fluoro-4-nitrophenol, chlorobenzene, reflux, 12 h; (d) $NH_3 \cdot H_2O$, acetone, 50 °C, 3 h; (e) DMF-DMA, CH_2Cl_2, reflux, 2–3 h; (f) CH_3NHNH_2, HOAC, 90 °C, 3 h; (g) $NH_2NH_2 \cdot H_2O$, EtOH, reflux, 3–6h; (h) **1a–b**, CH_2Cl_2, rt, 5 h.

Scheme 2 Reagents and conditions: (a) Pd/C, EtOH, rt, 4 h; (b) **1a–f**, CH_2Cl_2, rt, 4–7 h; (c) $NH_2NH_2 \cdot H_2O$, MeCN, reflux, 3–5 h; (d) DMF-DMA, CH_2Cl_2, reflux, 4–6 h; (e) appropriate amine, HOAc, 90 °C, 3h.

Table 1. Cellular antiproliferative activities of compounds **8a**, **8b**, **13a**, and **13b**.

Table 2. Cellular proliferative activities of compounds **13a–y**.

Table 3. Enzyme activities of compounds **13c–u** and **13x**.

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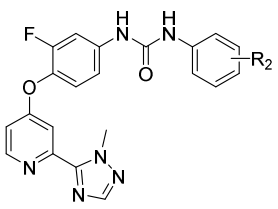
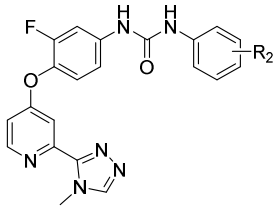
Table 1. Cellular antiproliferative activities of compounds **8a**, **8b**, **13a**, and **13b**.



8a and **8b**



13a and **13b**

Compd	R2	IC50 (μM)[a]			
		HT-29	H460	MDA-MB-231	WI-38
8a	3-CF_3-4-Cl	12.37 ± 2.38	NA	NA	NA
8b	3-CF_3	NA	11.62 ± 1.36	9.68 ± 0.52	22.37 ± 2.31
13a	3-CF_3-4-Cl	2.57 ± 0.31	2.31 ± 0.15	5.32 ± 0.06	9.85 ± 1.37
13b	3-CF_3	3.02 ± 0.08	1.27 ± 0.22	6.47 ± 0.37	12.62 ± 1.03
sorafenib		3.37 ± 0.38	2.25 ± 0.42	3.08 ± 0.50	8.42 ± 0.76

[a]The biological data are generated from at least three independent experiments.

NA: compound showing IC_{50} value > 50 μM.

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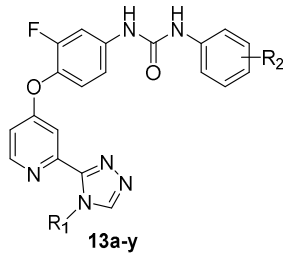
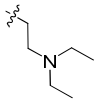
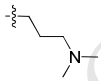
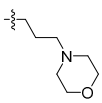
Table 2. Cellular proliferative activities of compounds **13a–y**.



13a-y

Compd	R1	R2	IC50 (μM)[a]				ClogD[b]
			HT-29	H460	MDA-MB-231	WI-38	
13a	CH_3	3-CF_3-4-Cl	2.57 ± 0.31	2.31 ± 0.15	5.32 ± 0.06	9.85 ± 1.37	4.58
13b	CH_3	3-CF_3	3.02 ± 0.08	1.27 ± 0.22	6.47 ± 0.37	12.62 ± 1.03	3.97
13c		3-CF_3-4-Cl	0.88 ± 0.05	1.15 ± 0.10	1.07 ± 0.03	3.76 ± 0.88	2.99
	T-1						
13d	T-1	3-CF_3	1.47 ± 0.06	2.15 ± 0.33	16.35 ± 3.28	11.22 ± 2.37	2.38
13e	T-1	2-CF_3	5.86 ± 1.27	NA	NA	NA	2.38
13f	T-1	3,4-$(F)_2$	3.62 ± 0.66	9.05 ± 2.28	NA	NA	1.79
13g	T-1	3-Cl-4-F	0.72 ± 0.11	1.58 ± 0.55	8.32 ± 1.43	7.58 ± 1.29	2.25
13h	T-1	2-4-$(Cl)_2$	1.89 ± 0.32	4.35 ± 0.62	7.96 ± 1.25	10.36 ± 0.67	2.71
13i		3-CF_3-4-Cl	0.90 ± 0.26	0.85 ± 0.32	1.54 ± 0.35	6.73 ± 0.79	3.18
	T-2						
13j	T-2	3-CF_3	0.63 ± 0.03	9.87 ± 1.75	0.72 ± 0.16	6.92 ± 1.34	2.57
13k	T-2	2-CF_3	3.41 ± 0.78	NA	11.56 ± 2.58	23.52 ± 0.68	2.57
13l	T-2	3,4-$(F)_2$	1.24 ± 0.53	14.85 ± 2.35	41.32 ± 3.73	NA	1.98
13m	T-2	3-Cl-4-F	0.50 ± 0.03	7.39 ± 1.30	34.00 ± 3.68	28.66 ± 3.53	2.44
13n	T-2	2-4-$(Cl)_2$	3.59 ± 0.76	5.38 ± 1.07	1.79 ± 0.33	4.28 ± 0.63	2.90
13o		3-CF_3-4-Cl	1.40 ± 0.26	1.68 ± 0.07	7.88 ± 0.72	7.91 ± 1.06	2.42
	T-3						
13p	T-3	3-CF_3	0.98 ± 0.19	1.79 ± 0.45	13.33 ± 2.38	11.55 ± 2.03	1.81
13q	T-3	2-CF_3	3.13 ± 1.08	10.86 ± 1.66	40.52 ± 3.76	28.62 ± 2.37	1.81
13r	T-3	3,4-$(F)_2$	1.21 ± 0.37	1.08 ± 0.29	25.25 ± 1.96	15.32 ± 1.56	1.22
13s	T-3	3-Cl-4-F	0.89 ± 0.12	1.27 ± 0.06	14.15 ± 1.28	5.37 ± 0.45	1.68
13t	T-3	2-4-$(Cl)_2$	0.68 ± 0.09	3.68 ± 0.37	7.49 ± 1.65	3.26 ± 0.68	2.14
13u		3-CF_3-4-Cl	2.86 ± 0.33	3.23 ± 0.17	8.28 ± 0.69	9.65 ± 1.33	4.09
	T-4						
13v	T-4	2-CF_3	15.70 ± 1.72	ND	ND	ND	3.49
13w	T-4	3-Cl-4-F	NA	16.23 ± 2.28	NA	ND	3.36

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13x	T-4	2-4-$(Cl)_2$	3.56 ± 0.35	ND	NA	ND	3.82
13y		3-CF_3-4-Cl	10.90 ± 2.32	NA	NA	ND	5.04
sorafenib			3.37 ± 0.38	2.25 ± 0.42	3.08 ± 0.50	8.42 ± 0.76	4.34

[a]The biological data are generated from at least three independent experiments.

[b]ClogD values, predicted at pH = 7.4, calculated using JChem_For_Excel_6.0.0.865.

NA: compound showing IC_{50} value > 50 μM.

ND: not determined.

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Table 3. Enzyme activities of compounds **13c–u** and **13x**.

Compd	% inhibition at 10 μM[a]				
	c-Kit	RET	FLT3	B-Raf	VEGFR-2
13c	70.4	74.1	66.3	15.7	0
13d	60.5	77.7	42.2	6.7	2.7
13e	22.7	43.0	17.6	0	2.2
13f	3.9	17.6	13.5	ND	ND
13g	74.0	63.3	52.2	0	26.9
13h	ND	ND	ND	0	6.0
13i	79.9	84.4	84.9	14.7	10.0
13j	66.5	72.1	61.0	3.3	11.5
13k	ND	ND	ND	ND	ND
13l	27.7	7.1	39.0	1.2	0
13m	40.4	1.9	59.6	0	2.7
13n	ND	ND	ND	ND	ND
13o	69.9	79.3	86.9	0	10.9
13p	69.9	85.6	69.2	11.8	62.8
13q	ND	ND	ND	24.8	1.7
13r	63.1	30.9	44.3	11.7	20.3
13s	71.0	45.5	72.1	27.4	35.0
13t	ND	ND	ND	0	11.3
13u	56.7	42.3	38.5	7.6	0
13x	43.5	45.6	22.0	ND	ND
sorafenib	95.6	97.0	98.5	100	96.8

[a]The biological data are means from at least two replicated experiments.

ND: not determined.

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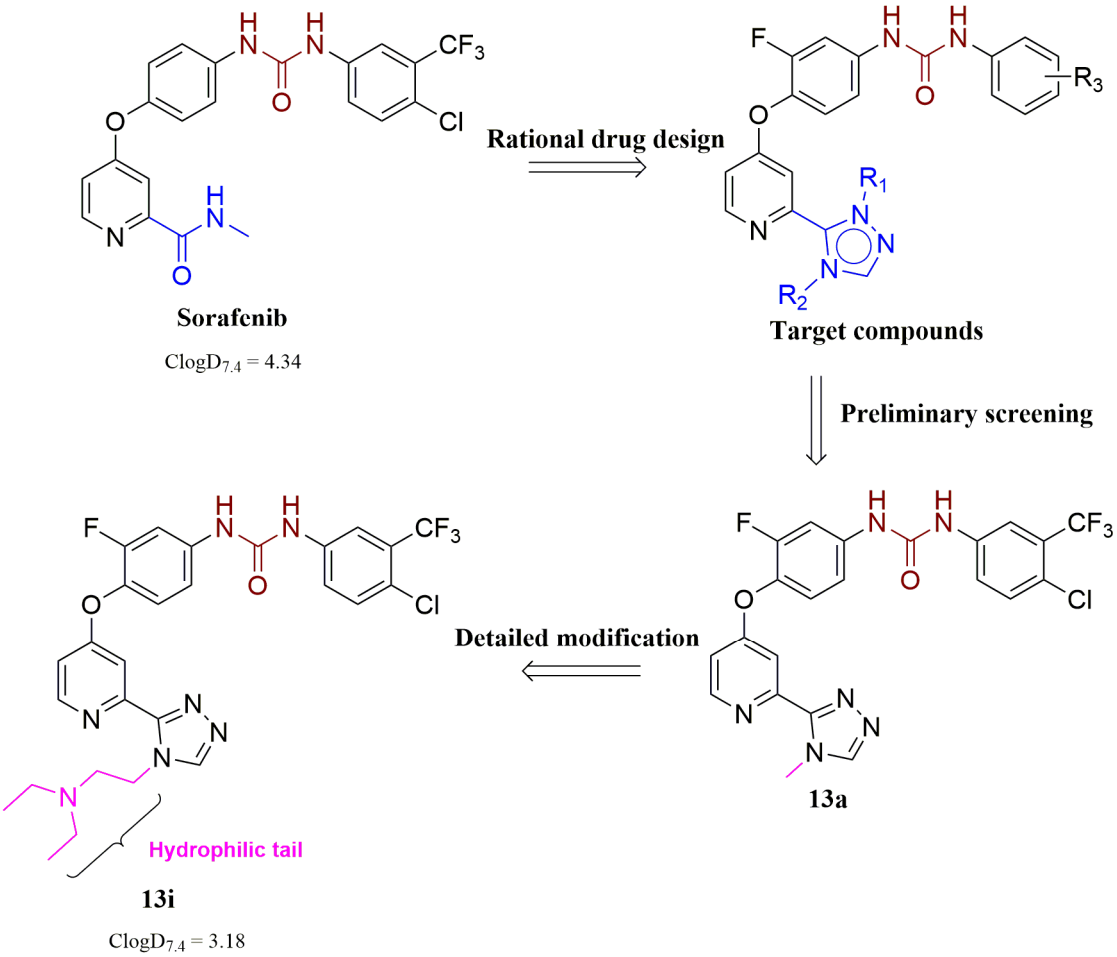
Rational drug design
Sorafenib
ClogD7.4 = 4.34
Target compounds
Preliminary screening
Detailed modification
13a
Hydrophilic tail
13i
ClogD7.4 = 3.18

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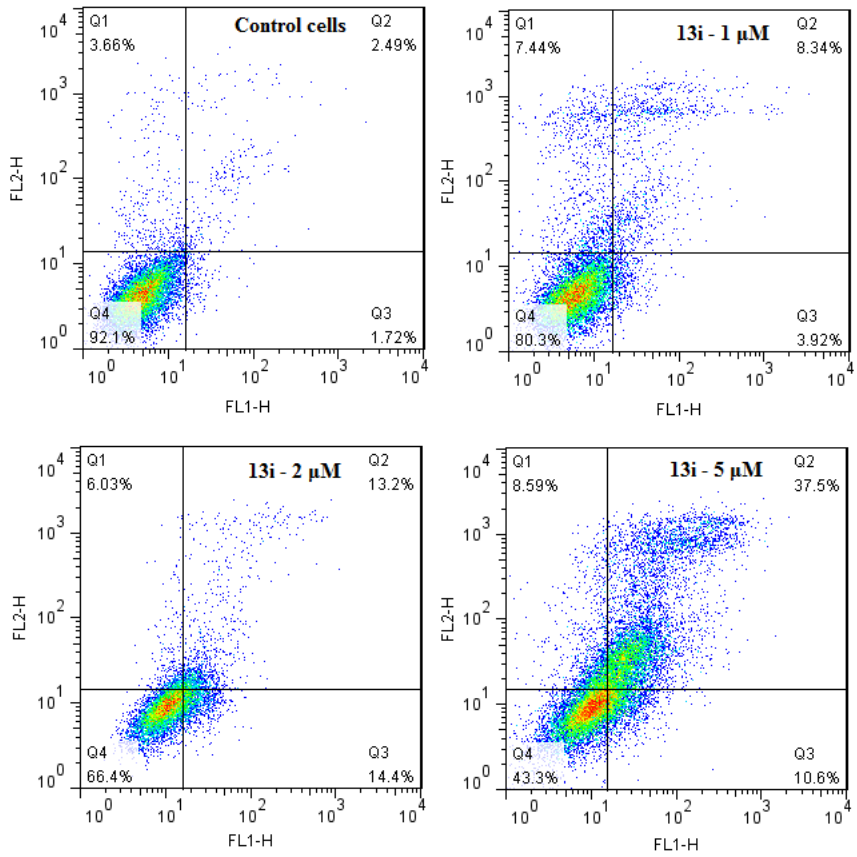

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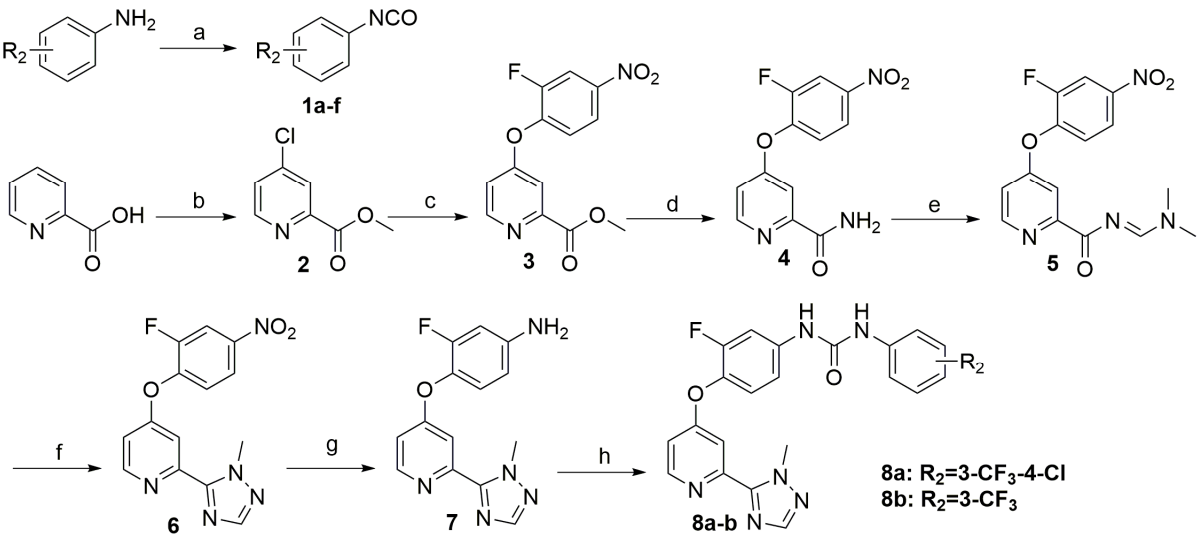

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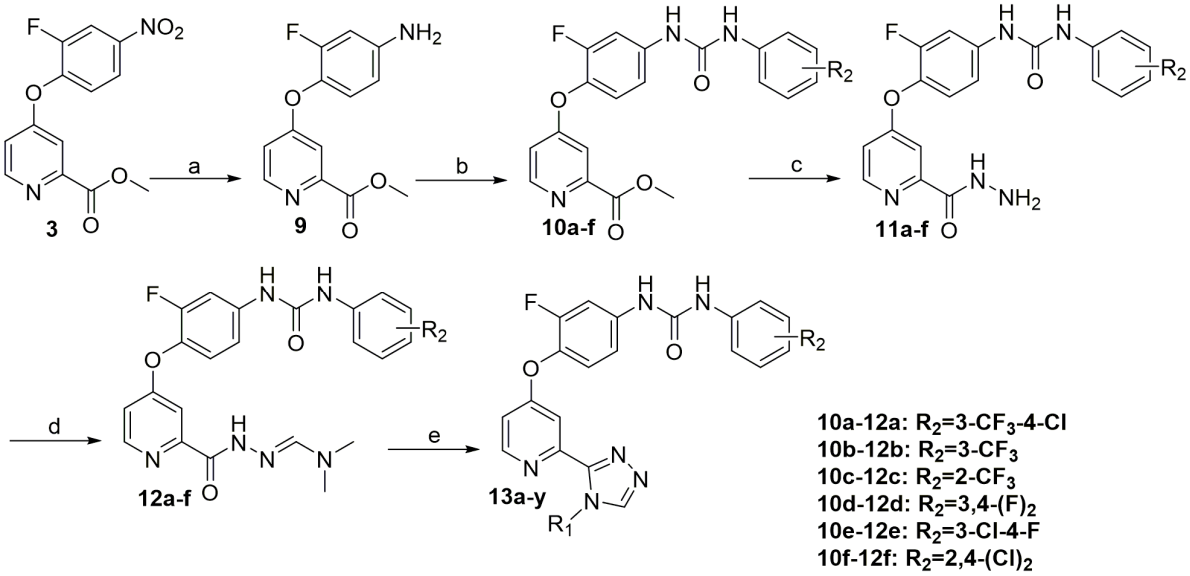

10a-12a: R_2=3-CF_3-4-Cl
10b-12b: R_2=3-CF_3
10c-12c: R_2=2-CF_3
10d-12d: R_2=3,4-$(F)_2$
10e-12e: R_2=3-Cl-4-F
10f-12f: R_2=2,4-$(Cl)_2$

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- A series of novel diaryl ureas were identified as potent antitumor agents.
- Compound **13i** potently suppressed proliferation of cancer cells.
- Compound **13i** effectively inhibited c-Kit, RET, and FLT3 kinases.
- Compounds **13i** significantly induced apoptosis of HT-29 cells.

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