Accepted Manuscript

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PII: S0223-5234(15)30052-0

DOI: 10.1016/j.ejmech.2015.05.025

Reference: EJMECH 7904

To appear in: *European Journal of Medicinal Chemistry*

Received Date: 31 October 2014

Revised Date: 9 May 2015

Accepted Date: 15 May 2015

Please cite this article as: X. Lv, H. Ying, X. Ma, N. Qiu, P. Wu, B. Yang, Y. Hu, Design, synthesis and biological evaluation of novel 4-alkynyl-quinoline derivatives as PI3K/mTOR dual inhibitors, European Journal of Medicinal Chemistry (2015), doi: 10.1016/j.ejmech.2015.05.025.

Graphic Abstract

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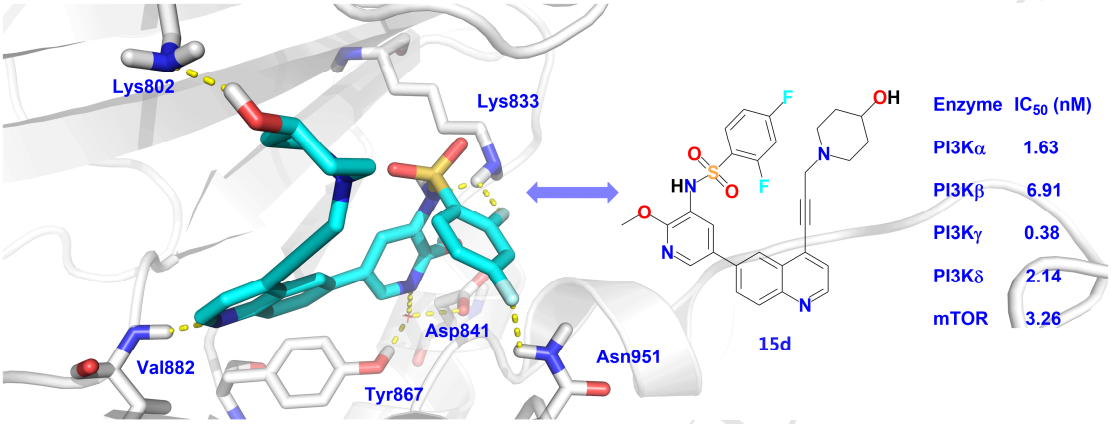

Design, synthesis and biological evaluation of novel 4-alkynyl-quinoline derivatives as PI3K/mTOR dual inhibitors

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Abstract: A novel series of 4-alkynyl-quinoline derivatives were designed, synthesized and biologically evaluated for their PI3Kα inhibitory activities and anti-proliferative effects against two cancer cell lines PC-3 and HCT-116. Most of them showed potent PI3Kα inhibitory activities with IC_{50} values at low nanomolar level and good to excellent anti-proliferative effects against both cell lines. Among them, compound **15d**, the most potent one, was selected for further biological evaluation. As a result, **15d** displayed strong inhibitory activity against other class I PI3K isoforms (PI3Kβ, PI3Kγ and PI3Kδ) and mTOR with an acceptable kinase selectivity profile. Moreover, the western blot assay indicated that the phosphorylation of Akt, another downstream effector of PI3K, can be remarkably suppressed by **15d** at cellular level. All these experimental results suggested that **15d** is a potent PI3K/mTOR dual inhibitor and could serve as a promising lead compound for the development of anticancer agents.

Keyword: PI3K, mTOR, Cancer, Inhibitor, 4-alkynyl-quinoline

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1. Introduction

The phosphoinositide 3-kinase (PI3K)/protein kinase B (PKB, also known as Akt)/mammalian target of rapamycin (mTOR) pathway is a key signal transduction system involved in the regulation of cell cycle progression, cell growth, survival and migration, and intracellular vesicular transport [1-3]. Accumulating studies have demonstrated that PI3K/Akt/mTOR signaling is frequently dysregulated in human cancers, resulting in constitutive activation of this signaling network [4-8]. Therefore, a sizeable effort has been devoted to development of cancer therapies targeting key kinase PI3K and/or downstream effectors such as Akt and mTOR [9-11]. In particular, dual inhibition of PI3K and mTOR has been considered as a more effective strategy for cancer therapy, since it can directly target the most commonly mutated kinase-PI3K [4-8] in this pathway as well as suppress PI3K-independent activation of mTOR. Meanwhile, dual inhibition of PI3K and mTOR avoids multiple mTOR-related negative feedback loops that a selective mTOR inhibitor usually fails to repress [12-15]. To date, a number of PI3K/mTOR dual inhibitors have been advanced into clinical trials [14, 16], such as GSK2126458 [17], BEZ235 [18, 19], BGT226 [20, 21], PF-04691502 [22], GDC-0980 [23] and PKI-587 [24], further suggesting that simultaneous inhibition of PI3K and mTOR has great potential for the treatment of cancer (**Fig. 1**).

<Insert **Fig. 1**>

In our previous work, a series of quinoxaline derivatives was identified to be PI3K inhibitors with favorable *in vitro* activities [25, 26]. However, the moderate *in vivo* efficacy of this class of compounds led us to explore the dual targeting strategies for an improved potency, especially the dual inhibition of PI3K and mTOR. Herein, we describe our initial efforts in this field to pursue the desired PI3K/mTOR dual inhibitors through structural modification of GSK2126458. As a PI3K/mTOR dual inhibitor under clinical trials, GSK2126458 displayed remarkable *in vitro* and *in vivo* potency, as well as excellent oral bioavailability [17]. Due to the availability of the co-crystal structure of GSK2126458 with PI3Kγ [17], we investigated their binding

mode seriously and envisioned that introduction of hydrophilic side chain onto C-4 position of the quinoline ring of GSK2126458 to replace pyridazine ring can not only improve the water solubility but also explore potential interactions with the residues located nearby ribose pocket (e.g. Lys802 and Ala805) (**Fig. 2**). In this study, an alkyne was employed as a linkage bridge between quinoline ring and hydrophilic group, affording a series of 4-alkynyl-quinoline derivatives (**Fig. 2**). All of them were subsequently prepared and evaluated for their *in vitro* PI3K inhibitory activities and anti-proliferative effects.

<Insert **Fig. 2**>

2. Results and discussion

2.1. Chemistry

The synthetic routes for 4-alkynyl-quinolines **15a**–**n**, **19a**–**e** are outlined in **Scheme 1**–**3**. Treatment of 5-bromo-2-chloro-3-nitropyridine **1** with sodium methoxide gave 5-bromo-2-methyloxy-3-nitropyridine **2**. Reduction of **2** with $SnCl_2$ in ethyl acetate produced the amine **3**. Sulfonylation of **3** with benzenesulfonyl chlorides gave corresponding sulfamides **4a**–**f**, which were then subjected to palladium-catalyzed Suzuki coupling with bis(pinacolato)diborane to afford boric acid esters **5a**–**f** (**Scheme 1**).

Condensation of 4-bromoaniline **6** with diethyl-2-(ethoxymethylene) malonate followed by intramolecular cyclization provided ethyl quinoline-3-carboxylate **8**. Following hydrolysis of **8**, the newly formed **9** was heated to remove the carboxylic group, leading to the generation of 4-hydroxyquinoline **10**. Treatment of **10** with $POCl_3$ afforded 4-chloroquinoline **11**, which was then treated with KI to give the 4-iodoquinoline **12**. Sonogashira coupling of **12** with alkynes **13a–l** in the presence of $Pd(PPh_3)_2Cl_2$ provided the corresponding intermediates **14a**–**l**. Coupling **14a**–**k** with **5a** via Suzuki reaction yielded corresponding target compounds **15a**–**k**. Similarly, compound **15l** was prepared by reaction of **16** with **5a** (**Scheme 2**).

Deprotection of the Boc-protected piperazidine derivative **14l** with TFA followed by acylation with acetyl chloride or methanesulfonyl chloride yielded **18a** and **18b**.

Subsequently, coupling **18a** and **18b** with **5a** afforded target compounds **15m** and **15n**, respectively. Compounds **19a**–**e** were obtained via the Suzuki coupling as described above (**Scheme 3**).

<Insert **Schemes 1-3**>

2.2. PI3Kα enzymatic and anti-proliferative assays

All synthesized target compounds were evaluated for their PI3Kα inhibitory activities and anti-proliferative activities against PC-3 and HCT-116 cell lines by Kinase-Glo Luminescent assay and Sulforhodamine B (SRB) assay, respectively. BEZ235 was used as the positive control. The experimental results of PI3Kα enzymatic assay summarized in **Table 1**, showed that all the tested 2, 4-difluorophenyl derivatives (**15a**–**n**) exhibited potent PI3Kα inhibitory activities with IC_{50} values in the range of 1.63–32.54 nM, better than that of the positive control BEZ235. However, compounds **19a**–**e** with mono-fluoro, methyl, trifluoromethyl or trifluoromethoxy on the phenyl showed moderate to weak inhibitory activities. These results suggested that the substitution pattern on the phenyl had a significant effect on inhibitory activity and the 2,4-difluoro substitutent on the phenyl appeared to be optimal in this series. On the other hand, among the 2,4-difluorophenyl derivatives (**15a**–**n**), compounds (**15a**, **15d**–**h**, **15m** and **15n**) with hydroxyl or amide group at the end of 4-alkynyl linker displayed more than a 4-fold improvement in the enzymatic activities compared to the compounds **15b** and **15i**–**k**. In addition, compound **15c** with *N*-methylpiperazine fragment and compound **15l** with no 4-alkynyl substituent merely exhibited moderate inhibitory activity among these derivatives.

According to the results of anti-proliferative assay (**Table 1**), the 4-alkynyl-quinoline derivatives showed moderate to potent activities and five compounds (**15d**, **15g**, **15h**, **15n** and **19b**) exhibited submicromolar potencies against PC-3 or HCT-116 cell line. Among them, it is noteworthy that compounds **15d** and **15g** with favorable values of molecular parameters (such as ClogP and tPSA) not only showed potent PI3Kα inhibitory potencies but also displayed excellent anti-proliferative activities. More encouragingly, 4-hydroxylpiperidine derivative **15d**,

the most potent compound in both the PI3K enzymatic and anti-proliferative assays, showed stronger activity against PC-3 cell line than that of BEZ235. Therefore, this compound can be used as a promising lead compound for further biological evaluation.

<Insert **Table 1**>

2.4. Kinase selectivity assay

In order to evaluate the activities of target compounds against other class I PI3Ks (PI3Kβ, PI3Kγ and PI3Kδ) and mTOR, compound **15d** was screened for its activities by ADP-Glo Luminescent assay and Lance Ultra assay, respectively. BEZ235 was used as the positive control. As shown in **Table 2**, compound **15d** displayed inhibitory activities with IC_{50} values ranging from single-digit nanomolar (PI3Kα, β, δ and mTOR) to subnanomolar (PI3Kγ), which were more potent than that of BEZ235. This result suggested that compound **15d** was a potent PI3K/mTOR dual inhibitor.

<Insert **Table 2**>

Subsequently, **15d** was submitted for screening in a panel of 50 kinases provided by DiscoveRx's KinomeScan service [27]. The results further confirmed that **15d** was a potent PI3K family and mTOR inhibitor showing moderate-to-good selectivity over other kinases with the exception of the PI4KIIIβ (*PIK4CB*) (**Table 3**).

<Insert **Table 3**>

2.5. Western blot assay

Finally, to determine whether **15d** suppressed the activation of Akt, a key node in the PI3K/Akt/mTOR signaling pathway, it was tested for its suppressive effect on pAkt(Ser473) level in PC-3 cells. Glyceraldehyde-3-phosphate dehydrogenase (GAPDH) was used as the internal control. The suppressive effect for **15d** was evaluated at the concentrations of 1 nM, 5 nM, 25 nM and 125 nM. As illustrated in **Fig. 4**, at the concentration of 125 nM, **15d** exhibited significantly inhibitory effect on

pAkt(Ser473), which further demonstrated **15d** can strongly down-regulate the PI3K/Akt/mTOR pathway.

<Insert **Fig. 3**>

2.3. Molecular docking study

To examine structure–activity relationships (SARs) in more detail, docking analysis of **15d** bound to PI3Kγ (PDB code 3L08), and **15d**, **19a** and **19b** bound to PI3Kα (PDB code 4JPS) was performed utilizing the Discovery Studio 2.1 software package, respectively. The result of **15d** bound to PI3Kγ as shown in **Fig. 4**, the nitrogen of sulfonamide in **15d** is involved in the hydrogen bonding interaction with Lys833 and the 4-hydroxy of the piperidine moiety at the end of 4-alkynyl linker forms an additional hydrogen bond with Lys802 near the ribose pocket. Additionally, a conserved water molecule is bridged between pyridyl nitrogen and residues Asp841 and Tyr867, and the nitrogen of quinoline makes a critical hydrogen bond with Val882 in the hinge region, which is consistent with the co-crystal structure of GSK2126458 bound to PI3Kγ. In addition, in the back pocket, the 2-fluoro and 4-fluoro on the phenyl moiety of **15d** form two hydrogen bonds with the residues Lys833 and Asn951, respectively. In contrast, the docking result of **15d** with PI3Kα showed the absence of one hydrogen bond between the 4-hydroxy of the piperidine moiety of **15d** and PI3Kα near the ribose pocket. In the other regions, **15d** forms five hydrogen bonds with residues (Lys802, Asp810, Tyr836, Val851 and Asn920) when bound to PI3Kα. These results were consistent with the molecule's (**15d**) potency and selectivity profiles (subnanomolar PI3Kγ potency and > 4-fold selectivity over PI3Kα).

The binding modes of **19a**, **19b** and **15d** bound to PI3Kα are generally identical. The difference lies in the interaction with the back pocket. In detail, 2-fluoro and 4-fluoro on the phenyl moiety of **15d** form two hydrogen bonds with the residues Lys802 and Asn920 in this region, respectively. However, as for **19a** and **19b**, only one hydrogen bond was formed between the 4-fluro or 3-fluro of the phenyl moiety

and the residue Asn920, thereby accounting for their significant loss in PI3Kα activities.

<Insert **Fig. 4**>

3. Conclusion

A novel series of 4-alkynyl-quinoline derivatives was evaluated as PI3K/mTOR dual inhibitors with potent enzymatic and cellular activities, and **15d** was identified as a potentially interesting lead molecule. Most of derivatives exhibited stronger PI3Kα inhibitory activities than that of BEZ235. Several compounds displayed potent anti-proliferative activities with IC_{50} values less than 1 μM, which were comparable to that of BEZ235. Additionally, the most potent compound **15d** with an acceptable kinase selectivity profile significantly inhibited other PI3Ks and mTOR, as well as the phosphorylation of pAkt(Ser473) at nanomolar level. According to these experimental results, it can be concluded that derivatization at the C-4 position of the quinoline core via introducing appropriate substituents to the alkynyl linker is a feasible way to attain promising inhibitors with high enzymatic and anti-proliferative activities.

4. Experimental section

4.1. Chemistry and chemical methods

^{1}H NMR and ^{13}C NMR spectra were recorded on the BRUKER AVII 400 (^{1}H: 400, ^{13}C: 100 MHz) and AVIII 500 (^{1}H: 500, ^{13}C: 125 MHz) NMR instruments. Chemical shifts are given in ppm (δ) relative to TMS as internal standard, coupling constants (J) are in hertz (Hz), and signals are using the following abbreviations: s, singlet; d, doublet; dd, doublet of doublets; t, triplet; td, doublet of triplets; q, quartet; m, multiplet, etc. Mass spectra (MS) were measured on an Esquire-LC-00075 spectrometer. Column chromatography and thin layer chromatography (TLC) were carried out using silica gel ZCX-3 and GF-254 (Qingdao haiyang chemical Co., Ltd.), respectively. Reagents and solvents were commercially available without further purification.

*4.1.1. 5-Bromo-2-methoxy-3-nitropyridine (**2**)*

To a solution of 5-bromo-2-chloro-3-nitropyridine (**1**) (10.0 g, 42.37 mmol) in anhydrous methanol (40 mL) was added sodium methoxide solution (20 mL, 63.56 mmol) slowly at 0 °C. The reaction mixture was then heated to 50 °C and stirred for 12 h. After the completion of reaction, the mixture was filtered and the precipitate was washed with water. The obtained solids were then dried under reduced pressure to give the title compound (8.12 g, 35.0 mmol, 83% yield) with light yellow. ESI-MS: m/z = 233 $[M+H]^+$.

*4.1.2. 5-Bromo-2-methoxypyridin-3-amine (**3**)*

A mixture of 5-bromo-2-methoxy-3-nitropyridine (**2**) (5.0 g, 21.55 mmol) and tin(II) chloride dihydrate (24.35 g, 107.75 mmol) was dissolved into ethyl acetate (0.2 L) and stirred at 50 °C for 3 h under a nitrogen atmosphere. The mixture was dissolved in EtOAc (0.8 L), washed with 1 N sodium hydroxide (1.2 L) twice, water twice, dried over magnesium sulfate and concentrated to give the title compound (3.31 g, 16.39 mmol, 76 % yield) as a brown solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 7.37 (d, J = 2.0 Hz, 1H, Ar-H), 6.97 (d, J = 2.0 Hz, 1H, Ar-H), 5.28 (s, 2H, NH_2), 3.82 (s, 3H, OCH_3). ESI-MS: m/z = 203 $[M+H]^+$.

*4.1.3. General procedure A for synthesis of N-(5-bromo-2-methoxypyridin-3-yl)benzenesulfonamides (**4a-f**)*

To a solution of 5-bromo-2-methoxypyridin-3-amine (**3**) (1.0 equiv) in anhydrous pyridine was added benzenesulfonyl chloride (1.0 equiv) at room temperature. The reaction was then stirred at room temperature for 24 h. The pyridine was removed under reduced pressure and the residue was purified by silica gel chromatography (10% ethyl acetate/petroleum ether to 100% ethyl acetate) to give the crude product. It was further washed with diethyl ether to give the title compound.

*4.1.3.1. N-(5-Bromo-2-methoxypyridin-3-yl)-2,4- difluorobenzenesulfonamide (**4a**)*

This compound was prepared from 5-bromo-2-methoxypyridin-3-amine (**3**) (3.0 g,

14.85 mmol) and 2,4-difluorobenzenesulfonyl chloride (3.15 g, 14.85 mmol) according to the general synthesis procedure A to afford the title compound (3.58 g, 9.47 mmol, 64 % yield) as a white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.41 (s, 1H, NH), 8.10 (d, *J* = 2.5 Hz, 1H, Ar-H), 7.75 (m, 1H, Ar-H), 7.74 (d, *J* = 2.5 Hz, 1H, Ar-H), 7.54 (td, *J* = 8.5, 2.0 Hz, 1H, Ar-H), 7.21 (td, *J* = 8.5, 2.0 Hz, 1H, Ar-H), 3.60 (s, 3H, OCH_3). ESI-MS: m/z = 379 $[M+H]^+$.

*4.1.3.2. N-(5-Bromo-2-methoxypyridin-3-yl)-4-fluorobenzenesulfonamide (**4b**)*

This compound was prepared from 5-bromo-2-methoxypyridin-3-amine (**3**) (3.0 g, 14.85 mmol) and 4-fluorobenzenesulfonyl chloride (2.88 g, 14.85 mmol) according to the general synthesis procedure A to afford the title compound (3.15 g, 8.75 mmol, 59% yield) as a white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.19 (s, 1H, NH), 8.08 (d, *J* = 2.5 Hz, 1H, Ar-H), 7.84 – 7.81 (m, 2H, Ar-H), 7.72 (d, *J* = 2.5 Hz, 1H, Ar-H), 7.43 (t, *J* = 8.5 Hz, 2H, Ar-H), 3.64 (s, 3H, OCH_3). ESI-MS: m/z = 361 $[M+H]^+$.

*4.1.3.3. N-(5-Bromo-2-methoxypyridin-3-yl)-3-fluorobenzenesulfonamide (**4c**)*

This compound was prepared from 5-bromo-2-methoxypyridin-3-amine (**3**) (3.0 g, 14.85 mmol) and 3-fluorobenzenesulfonyl chloride (2.88 g, 14.85 mmol) according to the general synthesis procedure A to afford the title compound (2.86 g, 7.94 mmol, 53% yield) as a white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.31 (s, 1H, NH), 8.09 (d, *J* = 2.5 Hz, 1H, Ar-H), 7.73 (d, *J* = 2.5 Hz, 1H, Ar-H), 7.65 (m, 1H, Ar-H), 7.59 (m, 2H, Ar-H), 7.55 (m, 1H, Ar-H), 3.64 (s, 3H, OCH_3). ESI-MS: m/z = 361 $[M+H]^+$.

*4.1.3.4. N-(5-Bromo-2-methoxypyridin-3-yl)-4-(trifluoromethyl)benzenesulfonamide (**4d**)*

This compound was prepared from 5-bromo-2-methoxypyridin-3-amine (**3**) (3.0 g, 14.85 mmol) and 4-(trifluoromethyl)benzene-1-sulfonyl chloride (3.62 g, 14.85 mmol) according to the general synthesis procedure A to afford the title compound (3.31 g,

8.07 mmol, 54% yield) as a white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.42 (s, 1H, NH), 8.11 (d, *J* = 2.5 Hz, 1H, Ar-H), 7.97 (q, *J* = 8.5 Hz, 4H, Ar-H), 7.77 (d, *J* = 2.5 Hz, 1H, Ar-H), 3.55 (s, 3H, OCH_3). ESI-MS: m/z = 411 $[M+H]^+$.

*4.1.3.5. N-(5-Bromo-2-methoxypyridin-3-yl)-4-(trifluoromethoxy)benzenesulfonamide (**4e**)*

This compound was prepared from 5-bromo-2-methoxypyridin-3-amine (**3**) (3.0 g, 14.85 mmol) and 4-(trifluoromethoxy)benzene-1-sulfonyl chloride (3.86 g, 14.85 mmol) according to the general synthesis procedure A to afford the title compound (3.55 g, 8.33 mmol, 56% yield) as a white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.28 (s, 1H, NH), 8.11 (d, *J* = 2.0 Hz, 1H, Ar-H), 7.86 (d, *J* = 8.5 Hz, 2H, Ar-H), 7.75 (d, *J* = 2.0 Hz, 1H, Ar-H), 7.59 (d, *J* = 8.5 Hz, 2H, Ar-H), 3.57 (s, 3H, OCH_3). ESI-MS: m/z = 427 $[M+H]^+$.

*4.1.3.6. N-(5-Bromo-2-methoxypyridin-3-yl)-4-methylbenzenesulfonamide (**4f**)*

This compound was prepared from 5-bromo-2-methoxypyridin-3-amine (**3**) (3.0 g, 14.85 mmol) and 4-methylbenzene-1-sulfonyl chloride (2.82 g, 14.85 mmol) according to the general synthesis procedure A to afford the title compound (3.02 g, 8.46 mmol, 57% yield) as a white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.07 (s, 1H, NH), 8.03 (d, *J* = 2.5 Hz, 1H, Ar-H), 7.68 (d, *J* = 8.0 Hz, 1H, Ar-H), 7.66 (d, *J* = 8.0 Hz, 2H, Ar-H), 7.38 (d, *J* = 8.0 Hz, 2H, Ar-H), 3.66 (s, 3H, OCH_3), 2.37 (s, 3H, CH_3). ESI-MS: m/z = 357 $[M+H]^+$.

*4.1.4. General procedure B for synthesis of N-(2-methoxy-5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)pyridin-3-yl)benzenesulfonamides (**5a-f**)*

To a two neck flask with a magnetic stir bar was added bis(pinacolato)diborane (1.0 equiv), *N*-(5-bromo-2-methoxypyridin-3-yl)benzenesulfonamide (1.0 equiv), $Pd(dppf)_2Cl_2$ (0.1 equiv), KOAc (3.0 equiv) and anhydrous dioxane. The mixture was deoxygenated by bubbling nitrogen through it for 10 min. The mixture was then

stirred at 100 °C for 3 h. After cooling to room temperature, the mixture was evaporated under reduced pressure and the residue was dissolved in EtOAc, washed with water twice and dried over magnesium sulfate. The crude product was purified by flash chromatography (25% ethyl acetate/petroleum ether) to give the target compound.

4.1.4.1.

*2,4-Difluoro-N-(2-methoxy-5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)pyridin-3-yl)benzenesulfonamide (**5a**)*

This compound was prepared from bis(pinacolato)diborane (335 mg, 1.32 mmol), *N*-(5-bromo-2-methoxypyridin-3-yl)-2,4-difluorobenzenesulfonamide (**4a**) (499 mg, 1.32 mmol), $Pd(dppf)_2Cl_2$ (97 mg, 0.13 mmol) and KOAc (388 mg, 3.96 mmol) according to the general synthesis procedure B to afford the title compound (495 mg, 1.16 mmol, 88% yield) as a white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.17 (s, 1H, NH), 8.20 (d, *J* = 1.5 Hz, 1H, Ar-H), 7.71 (d, *J* = 1.5 Hz, 1H, Ar-H), 7.69 (m, 1H, Ar-H), 7.56 (td, *J* = 8.5, 2.5 Hz, 1H, Ar-H), 7.19 (td, *J* = 8.5, 2.5 Hz, 1H, Ar-H), 3.62 (s, 3H, OCH_3), 1.29 (s, 12H, $CH_3 \times 4$). ESI-MS: m/z = 427 $[M+H]^+$.

4.1.4.2.

*4-Fluoro-N-(2-methoxy-5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)pyridin-3-yl)benzenesulfonamide (**5b**)*

This compound was prepared from bis(pinacolato)diborane (335 mg, 1.32 mmol), *N*-(5-bromo-2-methoxypyridin-3-yl)-4-fluorobenzenesulfonamide (**4b**) (475 mg, 1.32 mmol), $Pd(dppf)_2Cl_2$ (97 mg, 0.13 mmol) and KOAc (388 mg, 3.96 mmol) according to the general synthesis procedure B to afford the title compound (413 mg, 1.01 mmol, 77% yield) as a white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 9.91 (s, 1H, NH), 8.18 (d, *J* = 1.5 Hz, 1H, Ar-H), 7.75 – 7.71 (m, 3H, Ar-H), 7.40 (t, *J* = 8.5 Hz, 2H, Ar-H), 3.62 (d, *J* = 4.5 Hz, 3H, OCH_3), 1.30 (s, 12H, $CH_3 \times 4$). ESI-MS: m/z = 409 $[M+H]^+$.

4.1.4.3.

*3-Fluoro-N-(2-methoxy-5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)pyridin-3-yl)benzenesulfonamide (**5c**)*

This compound was prepared from bis(pinacolato)diborane (335 mg, 1.32 mmol), *N*-(5-bromo-2-methoxypyridin-3-yl)-3-fluorobenzenesulfonamide (**4c**) (475 mg, 1.32 mmol), $Pd(dppf)_2Cl_2$ (97 mg, 0.13 mmol) and KOAc (388 mg, 3.96 mmol) according to the general synthesis procedure B to afford the title compound (433 mg, 1.06 mmol, 80% yield) as a white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.06 (s, 1H, NH), 8.19 (d, *J* = 1.5 Hz, 1H, Ar-H), 7.73 (d, *J* = 1.5 Hz, 1H, Ar-H), 7.64 – 7.60 (m, 1H, Ar-H), 7.55-7.48 (m, 3H, Ar-H), 3.64(s, 3H, OCH_3), 1.30 (s, 12H, $CH_3 \times 4$). ESI-MS: m/z = 409 $[M+H]^+$.

4.1.4.4.

*N-(2-Methoxy-5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)pyridin-3-yl)-4-(trifluoromethyl)benzenesulfonamide (**5d**)*

This compound was prepared from bis(pinacolato)diborane (335 mg, 1.32 mmol), *N*-(5-bromo-2-methoxypyridin-3-yl)-4-(trifluoromethyl)benzenesulfonamide (**4d**) (541 mg, 1.32 mmol), $Pd(dppf)_2Cl_2$ (97 mg, 0.13 mmol) and KOAc (388 mg, 3.96 mmol) according to the general synthesis procedure B to afford the title compound (445 mg, 0.97 mmol, 73% yield) as a white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.15 (s, 1H, NH), 8.20 (d, *J* = 1.5 Hz, 1H, Ar-H), 7.97 (d, *J* = 8.5 Hz, 2H, Ar-H), 7.88 (d, *J* = 8.5 Hz, 2H, Ar-H), 7.69 (d, *J* = 1.5 Hz, 1H, Ar-H), 3.55 (s, 3H, OCH_3), 1.30 (s, 12H, $CH_3 \times 4$). ESI-MS: m/z = 459 $[M+H]^+$.

4.1.4.5.

*N-(2-Methoxy-5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)pyridin-3-yl)-4-(trifluoromethoxy)benzenesulfonamide (**5e**)*

This compound was prepared from bis(pinacolato)diborane (335 mg, 1.32 mmol), *N*-(5-bromo-2-methoxypyridin-3-yl)-4-(trifluoromethoxy)benzenesulfonamide (**4e**) (562 mg, 1.32 mmol), $Pd(dppf)_2Cl_2$ (97 mg, 0.13 mmol) and KOAc (388 mg, 3.96 mmol) according to the general synthesis procedure B to afford the title compound

(461 mg, 0.97 mmol, 73% yield) as a white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.01 (s, 1H, NH), 8.20 (d, *J* = 1.5 Hz, 1H, Ar-H), 7.79 (d, *J* = 8.5 Hz, 2H, Ar-H), 7.70 (d, *J* = 1.5 Hz, 1H, Ar-H), 7.57 (d, *J* = 8.5 Hz, 2H, Ar-H), 3.57 (s, 3H, OCH_3), 1.30 (s, 12H, CH_3 × 4). ESI-MS: m/z = 475 $[M+H]^+$.

4.1.4.6.

*N-(2-Methoxy-5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)pyridin-3-yl)-4-methylbenzenesulfonamide (**5f**)*

This compound was prepared from bis(pinacolato)diborane (335 mg, 1.32 mmol), *N*-(5-bromo-2-methoxypyridin-3-yl)-4-methylbenzenesulfonamide (**4f**) (470 mg, 1.32 mmol), $Pd(dppf)_2Cl_2$ (97 mg, 0.13 mmol) and KOAc (388 mg, 3.96 mmol) according to the general synthesis procedure B to afford the title compound (423 mg, 1.05 mmol, 80% yield) as a white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 9.76 (s, 1H, NH), 8.14 (d, *J* = 1.5 Hz, 1H, Ar-H), 7.73 (d, *J* = 1.5 Hz, 1H, Ar-H), 7.58 (d, *J* = 8.0 Hz, 2H, Ar-H), 7.35 (d, *J* = 8.0 Hz, 2H, Ar-H), 3.64 (s, 3H, OCH_3), 2.36 (s, 3H, CH_3), 1.30 (s, 12H, CH_3 × 4). ESI-MS: m/z = 405 $[M+H]^+$.

*4.1.5. Ethyl 6-bromo-4-hydroxyquinoline-3-carboxylate (**8**)*

A mixture of 4-bromoaniline (10.0 g, 58.48 mmol) and diethyl-2-(ethoxymethylene)malonate (12.63 g, 58.48 mmol) was stirred at 80 °C for 2 h. The mixture was evaporated under reduced pressure to give the diethyl 2-((4-bromophenylamino)methylene)malonate (**7**). This product was then treated with Ph_2O (100 mL) and stirred at 250 °C for 6 h. After cooling to 60 °C, petroleum ether (100 mL) was added to the mixture and the suspended solid was filtered, washed with petroleum ether, EtOAc and dried under reduced pressure to give the title compound (10.52 g, 35.66 mmol, 61% yield) as a white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 12.46 (s, 1H, OH), 8.60 (s, 1H, Ar-H), 8.23 (d, *J* = 2.5 Hz, 1H, Ar-H), 7.88 (dd, *J* = 8.5, 2.5 Hz, 1H, Ar-H), 7.61 (d, *J* = 8.5 Hz, 1H, Ar-H), 4.23 (q, *J* = 7.0 Hz, 2H, CH_2), 1.29 (t, *J* = 7.0 Hz, 3H, CH_3). ESI-MS: m/z = 296 $[M+H]^+$.

*4.1.6. 6-Bromo-4-hydroxyquinoline-3-carboxylic acid (**9**)*

Ethyl 6-bromo-4-hydroxyquinoline-3-carboxylate (**8**) (6.0 g, 20.34 mmol) and 2.5 N NaOH (50 mL) were charged in a 100 mL round-bottomed flask. The mixture was stirred at reflux for 1 h. After cooling to room temperature, the pH of the mixture was adjusted to 5 using 2 N HCl and the resulting solid was filtered and washed with water. The filter cake was then dried under reduced pressure to afford the title compound (5.15 g, 19.22 mmol, 94% yield) as a white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 12.51 (s, 1H), 8.63 (s, 1H, Ar-), 8.23 (d, J = 2.5 Hz, 1H, Ar-H), 7.87 (dd, J = 8.5, 2.5 Hz, 1H, Ar-H), 7.61 (d, J = 8.5 Hz, 1H, Ar-H). ESI-MS: m/z = 268 $[M+H]^+$.

*4.1.7. 6-Bromoquinolin-4-ol (**10**)*

6-Bromo-4-hydroxyquinoline-3-carboxylic acid (**9**) (5.0 g, 18.73 mmol) was added to a 100 mL round-bottomed flask in Ph_2O (30 mL) and then stirred at 260 °C for 2 h. After cooling to 60 °C, 30 mL petroleum ether was added and the suspended solid was filtered, washed with petroleum ether, EtOAc and dried under reduced pressure to give the title compound (3.21 g, 14.39 mmol, 77% yield) as a brown solid. ESI-MS: m/z = 224 $[M+H]^+$.

*4.1.8. 6-Bromo-4-chloroquinoline (**11**)*

To a 100 mL round-bottomed flask was added 6-bromoquinolin-4-ol (**10**) (3.0 g, 13.45 mmol), $POCl_3$ (20 mL) and DMF (0.5 mL). The mixture was stirred at reflux for 6 h. After cooling to room temperature, the reaction mixture was poured into ice water (100 mL) and stirred for 1 h. Then the pH of the mixture was adjusted to 8 using saturated aqueous $NaHCO_3$. The mixture was extracted with EtOAc and the organic phase was dried over sodium sulfate and concentrated in vacuo to give the title compound (2.75 g, 11.36 mmol, 84% yield) as a white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.87 (d, J = 4.5 Hz, 1H, Ar-H), 8.32 (d, J = 2.0 Hz, 1H, Ar-H), 8.03 (d, J = 9.0 Hz, 1H, Ar-H), 7.99 (dd, J = 9.0, 2.0 Hz, 1H, Ar-H), 7.82 (d, J = 4.5 Hz, 1H, Ar-H). ESI-MS: m/z = 242 $[M+H]^+$.

*4.1.9. 6-Bromo-4-iodoquinoline (**12**)*

To a solution of 6-bromo-4-chloroquinoline (**11**) (3.50 g, 14.46 mmol) in anhydrous EtOAc (20 mL) was added HCl-saturated EtOAc (40 mL) and a white precipitate formed immediately. After stirring for 30 min, the suspension was concentrated under vacuum to afford 6-bromo-4-chloroquinoline hydrochloride as an off white solid (3.91 g, 14.14 mmol).

A two-neck flask was charged with 6-bromo-4-chloroquinoline hydrochloride (3.91 g, 14.14mmol), anhydrous potassium iodide (9.76 g, 70.70 mmol) and anhydrous acetonitrile (100 mL). The resulting slurry was stirred at reflux for 48 h and allowed to cool to room temperature. Saturated aqueous $NaHCO_3$ solution (40 mL) was added to the mixture, followed by 20 mL of a 5% sodium sulfite solution. The reaction mixture was extracted with CH_2Cl_2 (200 mL × 2). The combined organic extracts were dried over magnesium sulfate and concentrated in vacuo to give the crude product, which was further purified by silica gel column chromatography (25% ethyl acetate/petroleum ether) to give the title compound (4.42 g, 13.27 mmol, 94% yield) as an off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.51 (d, J = 4.5 Hz, 1H, Ar-H), 8.21 (d, J = 4.5 Hz, 1H, Ar-H), 8.11 (t, J = 1.5 Hz, 1H, Ar-H), 7.97 – 7.91 (m, 2H, Ar-H). ESI-MS: m/z = 334 $[M+H]^+$.

*4.1.10. General procedure C for synthesis of aliphatic propargylamines (**13b-h** and **13l**)*

A solution of 3-bromopropyne (1.0 equiv), amine (1.0 equiv) and K_2CO_3 (2.0 equiv) in THF was stirred at reflux for 6 h. The solvent was removed and the residue was partitioned between water and CH_2Cl_2. The organic layer was dried over magnesium sulfate and under reduced pressure. The crude product was further purified by silica gel column chromatography to give the desired propargylamine.

*4.1.10.1. 1-(Prop-2-ynyl)piperidine (**13b**)*

This compound was prepared from 3-bromopropyne (500 mg, 4.24 mmol), piperidine (360 mg, 4.24 mmol) and K_2CO_3 (1.17 g, 8.48 mmol) according to the general synthesis procedure C to afford the title compound (344 mg, 2.80 mmol, 66% yield) as a viscous oil. ESI-MS: m/z = 124 $[M+H]^+$.

*4.1.10.2. 1-Methyl-4-(prop-2-ynyl)piperazine (**13c**)*

This compound was prepared from 3-bromopropyne (500 mg, 4.24 mmol), *N*-methylpiperazine (424 mg, 4.24 mmol) and K_2CO_3 (1.17 g, 8.48 mmol) according to the general synthesis procedure C to afford the title compound (359 mg, 2.59 mmol, 61% yield) as a white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 3.24 (d, J = 2.5 Hz, 2H, CH_2), 3.14 (t, J = 2.5 Hz, 1H, acetylenic hydrogen), 2.44 (s, 4H, CH_2 × 2), 2.30 (s, 4H, CH_2 × 2), 2.15 (s, 3H, CH_3). ESI-MS: m/z = 139 $[M+H]^+$.

*4.1.10.3. 1-(Prop-2-ynyl)piperidin-4-ol (**13d**)*

This compound was prepared from 3-bromopropyne (500 mg, 4.24 mmol), 4-hydroxypiperidine (428 mg, 4.24 mmol) and K_2CO_3 (1.17 g, 8.48 mmol) according to general synthesis procedure C to afford the title compound (403 mg, 2.90 mmol, 68% yield) as a white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 4.53 (d, J = 3.5 Hz, 1H, OH), 3.48 – 3.31 (m, 1H, CH), 3.19 (d, J = 2.0 Hz, 2H, CH_2), 3.09 (d, J = 2.0 Hz, 1H, acetylenic hydrogen), 2.69 – 2.59 (m, 2H, CH_2), 2.14 (t, J = 9.5 Hz, 2H, CH_2), 1.68 (d, J = 9.5 Hz, 2H, CH_2), 1.41 – 1.30 (m, 2H, CH_2). ESI-MS: m/z = 140 $[M+H]^+$.

*4.1.10.4. 1-(Prop-2-ynyl)piperidin-3-ol (**13e**)*

This compound was prepared from 3-bromopropyne (500 mg, 4.24 mmol), 3-hydroxypiperidine (428 mg, 4.24 mmol) and K_2CO_3 (1.17 g, 8.48 mmol) according to general synthesis procedure C to afford the title compound (392 mg, 2.82 mmol, 67% yield) as a white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 4.60 (d, J = 5.0 Hz, 1H, OH), 3.43 (m, 1H, CH), 3.21 (d, J = 2.0 Hz, 2H, CH_2), 3.10 (d, J = 2.0 Hz, 1H, acetylenic hydrogen), 2.74 (m, 1H, CH_2), 2.55 (m, 1H, CH_2), 1.98 (m, 1H, CH_2), 1.86 (m, 1H, CH_2), 1.74 (m, 1H, CH_2), 1.64 – 1.56 (m, 1H, CH_2), 1.42 – 1.32 (m, 1H, CH_2),

0.99 (m, 1H, CH_2). ESI-MS: m/z = 140 $[M+H]^+$.

*4.1.10.5. 2-(Methyl(prop-2-ynyl)amino)ethanol (**13g**)*

This compound was prepared from 3-bromopropyne (500 mg, 4.24 mmol), 2-methylaminoethanol (318 mg, 4.24 mmol) and K_2CO_3 (1.17 g, 8.48 mmol) according to the general synthesis procedure C to afford the title compound (271 mg, 2.40 mmol, 57% yield) as a viscous oil. ^{1}H NMR (500 MHz, DMSO-d_6) δ 4.42 (s, 1H, OH), 3.45 (t, *J* = 6.5 Hz, 2H, CH_2), 3.30 (d, *J* = 2.5 Hz, 2H, CH_2), 3.10 (t, *J* = 2.5 Hz, 1H, acetylenic hydrogen), 2.43 (t, *J* = 6.5 Hz, 2H, CH_2), 2.21 (s, 3H, CH_3). ESI-MS: m/z = 114 $[M+H]^+$.

*4.1.10.6. 3-(Methyl(prop-2-ynyl)amino)propane-1,2-diol (**13h**)*

This compound was prepared from 3-bromopropyne (500 mg, 4.24 mmol), 3-methylamino-1,2-propanediol (445 mg, 4.24 mmol) and K_2CO_3 (1.17 g, 8.48 mmol) according to general synthesis procedure C to afford the title compound (314 mg, 2.18 mmol, 51% yield) as a viscous oil. ^{1}H NMR (500 MHz, DMSO-d_6) δ 4.48 (s, 1H, OH), 4.41 (d, *J* = 4.5 Hz, 1H, OH), 3.54 (m, 1H, CH), 3.34 – 3.27 (m, 4H, CH_2 × 2), 3.10 (t, *J* = 2.5 Hz, 1H, acetylenic hydrogen), 2.40 (dd, *J* = 12.5, 5.0 Hz, 1H, CH_2), 2.28 (dd, *J* = 12.5, 7.0 Hz, 1H, CH_2), 2.23 (s, 3H, CH_3). ESI-MS: m/z = 144 $[M+H]^+$.

*4.1.10.7. Tert-butyl 4-(prop-2-ynyl)piperazine-1-carboxylate (**13l**)*

This compound was prepared from 3-bromopropyne (500 mg, 4.24 mmol), *N*-(tert-butoxycarbonyl)piperazine (789 mg, 4.24 mmol) and K_2CO_3 (1.17 g, 8.48 mmol) according to the general synthesis procedure C to afford the title compound (515 mg, 2.30 mmol, 54% yield) as a viscous oil. ^{1}H NMR (500 MHz, DMSO-d_6) δ 3.33 (m, 4H, CH_2 × 2), 3.29 (d, *J* = 2.5 Hz, 2H, CH_2), 3.18 (t, *J* = 2.5 Hz, 1H, acetylenic hydrogen), 2.42 – 2.33 (m, 4H, CH_2 × 2), 1.40 (s, 9H, CH_3 × 3). ESI-MS: m/z = 225 $[M+H]^+$.

*4.1.11. General procedure D for synthesis of aromatic propargylamines (**13i-k**)*

To a round-bottomed flask was added 3-bromopropyne (1.0 equiv), phenylamine (1.0 equiv), K_2CO_3 (2.0 equiv) and acetone. The mixture was stirred at room temperature for 24 h. The solvent was removed and the residue was partitioned between water and CH_2Cl_2. The organic layer was dried over magnesium sulfate and under reduced pressure. The crude product was further purified by silica gel column chromatography to give the desired propargylamine.

*4.1.11.1. N-(Prop-2-ynyl)aniline (**13i**)*

This compound was prepared from 3-bromopropyne (500 mg, 4.24 mmol), aniline (394 mg, 4.24 mmol) and K_2CO_3 (1.17 g, 8.48 mmol) according to the general synthesis procedure D to afford the title compound (183 mg, 1.39 mmol, 33% yield) as a viscous oil. ^{1}H NMR (500 MHz, DMSO-d_6) δ 7.11 (dd, J = 8.5, 7.5 Hz, 2H, Ar-H), 6.64 (d, J = 8.5 Hz, 2H, Ar-H), 6.60 (t, J = 7.5 Hz, 1H, Ar-H), 5.94 (t, J = 6.0 Hz, 1H, NH), 3.85 (dd, J = 6.0, 2.5 Hz, 2H, CH_2), 3.05 (t, J = 2.5 Hz, 1H, acetylenic hydrogen). ESI-MS: m/z = 132 $[M+H]^+$.

*4.1.11.2. 4-Methoxy-N-(prop-2-ynyl)aniline (**13j**)*

This compound was prepared from 3-bromopropyne (500 mg, 4.24 mmol), p-anisidine (522 mg, 4.24 mmol) and K_2CO_3 (1.17 g, 8.48 mmol) according to the general synthesis procedure D to afford the title compound (151 mg, 0.94 mmol, 22% yield) as a viscous oil. ^{1}H NMR (500 MHz, DMSO-d_6) δ 6.75 (d, J = 9.0 Hz, 2H, Ar-H), 6.61 (d, J = 9.0 Hz, 2H, Ar-H), 5.52 (t, J = 6.5 Hz, 1H, NH), 3.80 (dd, J = 6.5, 2.5 Hz, 2H, CH_2), 3.65 (s, 3H, OCH_3), 3.02 (t, J = 2.5 Hz, 1H, acetylenic hydrogen). ESI-MS: m/z = 162 $[M+H]^+$.

*4.1.11.3. 4-Fluoro-N-(prop-2-ynyl)aniline (**13k**)*

This compound was prepared from 3-bromopropyne (500 mg, 4.24 mmol), 4-fluoroaniline (471 mg, 4.24 mmol) and K_2CO_3 (1.17 g, 8.48 mmol) according to the general synthesis procedure D to afford the title compound (179 mg, 1.20 mmol, 28% yield) as a viscous oil. ^{1}H NMR (500 MHz, DMSO-d_6) δ 6.96 (t, J = 9.0 Hz, 2H,

Ar-H), 6.65 – 6.61 (m, 2H, Ar-H), 5.90 (t, *J* = 6.0 Hz, 1H, NH), 3.84 (dd, *J* = 6.0, 2.5 Hz, 2H, CH_2), 3.05 (t, *J* = 2.5 Hz, 1H, acetylenic hydrogen). ESI-MS: m/z = 150 $[M+H]^+$.

*4.1.12. General procedure E for sonogashira coupling reaction (**14a-l**)*

6-Bromo-4-iodoquinoline (**12**) (1.0 equiv), $Pd(PPh_3)_2Cl_2$ (0.1 equiv), CuI (0.15 equiv) and triethylamine were charged in a three-neck round bottom flask. The flask was fitted with a N_2 inlet adaptor and purged with N_2 for 10 min. The solution of alkyne (1.0 equiv) was then added via syringe and purged with N_2 for another 10 min. The reaction mixture was stirred at 50 °C for 5 h. After the completion of reaction, the mixture was concentrated under reduced pressure and the residue was dissolved in EtOAc, washed with 1 N NaOH and water, then the organic phase was dried over magnesium sulfate. The crude product was purified by silica gel column chromatography yielded the desired compound.

*4.1.12.1. 4-(6-Bromoquinolin-4-yl)but-3-yn-1-ol (**14a**)*

This compound was prepared from 6-bromo-4-iodoquinoline (**12**) (100 mg, 0.30 mmol) and commercially available but-3-yn-1-ol (**13a**) (21 mg, 0.30 mmol) according to the general synthesis procedure E to afford the title compound (58 mg, 0.21 mmol, 70% yield) as an off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.88 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.38 (d, *J* = 2.0 Hz, 1H, Ar-H), 7.98 (d, *J* = 9.0 Hz, 1H, Ar-H), 7.93 (dd, *J* = 9.0, 2.0 Hz, 1H, Ar-H), 7.61 (d, *J* = 4.5 Hz, 1H, Ar-H), 5.05 (t, *J* = 5.5 Hz, 1H, OH), 3.69 (q, *J* = 6.5 Hz, 2H, CH_2), 2.77 (t, *J* = 6.5 Hz, 2H, CH_2). ESI-MS: m/z = 276 $[M+H]^+$.

*4.1.12.2. 6-Bromo-4-(3-(piperidin-1-yl)prop-1-ynyl)quinoline (**14b**)*

This compound was prepared from 6-bromo-4-iodoquinoline (**12**) (100 mg, 0.30 mmol) and 1-(prop-2-ynyl)piperidine (**13b**) (37 mg, 0.30 mmol) according to the general synthesis procedure E to afford the title compound (56 mg, 0.17 mmol, 57% yield) as an off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.90 (d, *J* = 4.5 Hz, 1H,

Ar-H), 8.38 (d, *J* = 2.0 Hz, 1H, Ar-H), 8.00 (d, *J* = 9.0 Hz, 1H, Ar-H), 7.94 (dd, *J* = 9.0, 2.0 Hz, 1H, Ar-H), 7.66 (d, *J* = 4.5 Hz, 1H, Ar-H), 3.70 (s, 2H, CH_2), 2.56 (m, 4H, CH_2 × 2), 1.58 – 1.52 (m, 4H, CH_2 × 2), 1.42 – 1.37 (m, 2H, CH_2). ESI-MS: m/z = 329 $[M+H]^+$.

*4.1.12.3. 6-Bromo-4-(3-(4-methylpiperazin-1-yl)prop-1-ynyl)quinoline (**14c**)*

This compound was prepared from 6-bromo-4-iodoquinoline (**12**) (100 mg, 0.30 mmol) and 1-methyl-4-(prop-2-ynyl)piperazine (**13c**) (41 mg, 0.30 mmol) according to the general synthesis procedure E to afford the title compound (62 mg, 0.18 mmol, 60% yield) as a viscous oil. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.87 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.36 (d, *J* = 2.0 Hz, 1H, Ar-H), 7.98 (d, *J* = 9.0 Hz, 1H, Ar-H), 7.80 (dd, *J* = 9.0, 2.0 Hz, 1H, Ar-H), 7.61 (d, *J* = 4.5 Hz, 1H, Ar-H), 3.76 (s, 2H, CH_2), 3.01 – 2.85 (m, 8H, CH_2 × 4), 2.57 (s, 3H, CH_3). ESI-MS: m/z = 344 $[M+H]^+$.

*4.1.12.4. 1-(3-(6-Bromoquinolin-4-yl)prop-2-ynyl)piperidin-4-ol (**14d**)*

This compound was prepared from 6-bromo-4-iodoquinoline (**12**) (100 mg, 0.30 mmol) and 1-(prop-2-ynyl)piperidin-4-ol (**13d**) (42 mg, 0.30 mmol) according to the general synthesis procedure E to afford the title compound (58 mg, 0.17 mmol, 57% yield) as an off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.92 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.38 (d, *J* = 2.0 Hz, 1H, Ar-H), 8.03 (d, *J* = 9.0 Hz, 1H, Ar-H), 7.97 (dd, *J* = 9.0, 2.0 Hz, 1H, Ar-H), 7.68 (d, *J* = 4.5 Hz, 1H, Ar-H), 4.61 (d, *J* = 4.5 Hz, 1H, OH), 3.74 (s, 2H, CH_2), 3.50 (m, 1H, CH), 2.86 (m, 2H, CH_2), 2.44 – 2.30 (m, 2H, CH_2), 1.77 (m, 2H, CH_2), 1.47 (m, 2H, CH_2). ESI-MS: m/z = 345 $[M+H]^+$.

*4.1.12.5. 1-(3-(6-Bromoquinolin-4-yl)prop-2-ynyl)piperidin-3-ol (**14e**)*

This compound was prepared from 6-bromo-4-iodoquinoline (**12**) (100 mg, 0.30 mmol) and 1-(prop-2-ynyl)piperidin-3-ol (**13e**) (42 mg, 0.30 mmol) according to the general synthesis procedure E to afford the title compound (51 mg, 0.15 mmol, 50% yield) as an off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.92 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.38 (d, *J* = 2.0 Hz, 1H, Ar-H), 8.02 (d, *J* = 9.0 Hz, 1H, Ar-H), 7.96 (dd, *J* =

9.0, 2.0 Hz, 1H, Ar-H), 7.67 (d, *J* = 4.5 Hz, 1H, Ar-H), 4.72 (s, 1H, OH), 3.76 (s, 2H, CH_2), 3.56 (m, 1H, CH), 2.94 (dd, *J* = 10.0, 4.0 Hz, 1H, CH_2), 2.77 (d, *J* = 11.0 Hz, 1H, CH_2), 2.24 (td, *J* = 11.0, 3.0 Hz, 1H, CH_2), 2.10 (t, *J* = 10.0 Hz, 1H, CH_2), 1.85 – 1.79 (m, 1H, CH_2), 1.74 – 1.67 (m, 1H, CH_2), 1.52 – 1.44 (m, 1H, CH_2), 1.15 – 1.07 (m, 1H, CH_2). ESI-MS: m/z = 345 $[M+H]^+$.

*4.1.12.6. 2-(3-(6-Bromoquinolin-4-yl)prop-2-ynyloxy)ethanol (**14f**)*

This compound was prepared from 6-bromo-4-iodoquinoline (**12**) (100 mg, 0.30 mmol) and commercially available 2-(prop-2-ynyloxy)ethanol (**13f**) (30 mg, 0.30 mmol) according to the general synthesis procedure E to afford the title compound (55 mg, 0.18 mmol, 60% yield) as an off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.92 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.32 (d, *J* = 2.0 Hz, 1H, Ar-H), 8.01 (d, *J* = 9.0 Hz, 1H, Ar-H), 7.95 (dd, *J* = 9.0, 2.0 Hz, 1H, Ar-H), 7.69 (d, *J* = 4.5 Hz, 1H, Ar-H), 4.73 (t, *J* = 5.5 Hz, 1H, OH), 4.61 (s, 2H, CH_2), 3.64 – 3.61 (m, 2H, CH_2), 3.60 – 3.56 (m, 2H, CH_2). ESI-MS: m/z = 306 $[M+H]^+$.

*4.1.12.7. 2-((3-(6-Bromoquinolin-4-yl)prop-2-ynyl)(methyl)amino)ethanol (**14g**)*

This compound was prepared from 6-bromo-4-iodoquinoline (**12**) (100 mg, 0.30 mmol) and 2-(methyl(prop-2-ynyl)amino)ethanol (**13g**) (34 mg, 0.30 mmol) according to the general synthesis procedure E to afford the title compound (51 mg, 0.16 mmol, 53% yield) as a viscous oil. ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.90 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.36 (d, *J* = 2.0 Hz, 1H, Ar-H), 8.00 (d, *J* = 9.0 Hz, 1H, Ar-H), 7.95 (dd, *J* = 9.0, 2.0 Hz, 1H, Ar-H), 7.66 (d, *J* = 4.5 Hz, 1H, Ar-H), 4.49 (t, *J* = 5.5 Hz, 1H, OH), 3.78 (s, 2H, CH_2), 3.53 (q, *J* = 6.0 Hz, 2H, CH_2), 2.58 (t, *J* = 6.0 Hz, 2H, CH_2), 2.37 (s, 3H, CH_3). ESI-MS: m/z = 319 $[M+H]^+$.

*4.1.12.8. 3-((3-(6-Bromoquinolin-4-yl)prop-2-ynyl)(methyl)amino)propane-1,2-diol (**14h**)*

This compound was prepared from 6-bromo-4-iodoquinoline (**12**) (100 mg, 0.30 mmol) and 3-(methyl(prop-2-ynyl)amino)propane-1,2-diol (**13h**) (43 mg, 0.30 mmol)

according to the general synthesis procedure E to afford the title compound (66 mg, 0.19 mmol, 63% yield) as a viscous oil. ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.90 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.37 (d, *J* = 2.0 Hz, 1H, Ar-H), 8.00 (d, *J* = 9.0 Hz, 1H, Ar-H), 7.94 (dd, *J* = 9.0, 2.0 Hz, 1H, Ar-H), 7.66 (d, *J* = 4.5 Hz, 1H, Ar-H), 4.50 (d, *J* = 4.5 Hz, 2H, OH × 2), 3.80 (s, 2H, CH_2), 3.62 (m, 1H, CH), 3.40 – 3.33 (m, 2H, CH_2), 2.57 (dd, *J* = 12.5, 5.0 Hz, 1H, CH_2), 2.44 (dd, *J* = 12.5, 7.0 Hz, 1H, CH_2), 2.39 (s, 3H, CH_3). ESI-MS: m/z = 349 $[M+H]^+$.

*4.1.12.9. N-(3-(6-Bromoquinolin-4-yl)prop-2-ynyl)aniline (**14i**)*

This compound was prepared from 6-bromo-4-iodoquinoline (**12**) (100 mg, 0.30 mmol) and *N*-(prop-2-ynyl)aniline (**13i**) (39 mg, 0.30 mmol) according to the general synthesis procedure E to afford the title compound (60 mg, 0.18 mmol, 60% yield) as a viscous oil. ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.89 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.24 (d, *J* = 2.0 Hz, 1H, Ar-H), 7.98 (d, *J* = 9.0 Hz, 1H, Ar-H), 7.92 (dd, *J* = 9.0, 2.0 Hz, 1H, Ar-H), 7.60 (d, *J* = 4.5 Hz, 1H, Ar-H), 7.19 (dd, *J* = 9.5, 9.0 Hz, 2H, Ar-H), 6.80 (d, *J* = 9.5 Hz, 2H, Ar-H), 6.66 (t, *J* = 9.0 Hz, 1H, Ar-H), 6.21 (t, *J* = 6.5 Hz, 1H, NH), 4.35 (d, *J* = 6.5 Hz, 2H, CH_2). ESI-MS: m/z = 337 $[M+H]^+$.

*4.1.12.10. N-(3-(6-Bromoquinolin-4-yl)prop-2-ynyl)-4-methoxyaniline (**14j**)*

This compound was prepared from 6-bromo-4-iodoquinoline (**12**) (100 mg, 0.30 mmol) and 4-methoxy-*N*-(prop-2-ynyl)aniline (**13j**) (48 mg, 0.30 mmol) according to the general synthesis procedure E to afford the title compound (52 mg, 0.14 mmol, 47% yield) as a viscous oil. ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.87 (s, 1H, Ar-H), 8.16 (s, 1H, Ar-H), 7.95 (d, *J* = 9.0 Hz, 1H, Ar-H), 7.89 (d, *J* = 9.0 Hz, 1H, Ar-H), 7.57 (d, *J* = 4.0 Hz, 1H, Ar-H), 6.80 (d, *J* = 8.5 Hz, 2H, Ar-H), 6.75 (d, *J* = 8.5 Hz, 2H, Ar-H), 5.79 (t, *J* = 6.5 Hz, 1H, NH), 4.27 (d, *J* = 6.5 Hz, 2H, CH_2), 3.64 (s, 3H, OCH_3). ESI-MS: m/z = 367 $[M+H]^+$.

*4.1.12.11. N-(3-(6-Bromoquinolin-4-yl)prop-2-ynyl)-4-fluoroaniline (**14k**)*

This compound was prepared from 6-bromo-4-iodoquinoline (**12**) (100 mg, 0.30 mmol) and 4-fluoro-*N*-(prop-2-ynyl)aniline (**13k**) (45 mg, 0.30 mmol) according to the general synthesis procedure E to afford the title compound (63 mg, 0.18 mmol, 60% yield) as a viscous oil. ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.87 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.13 (d, *J* = 2.0 Hz, 1H, Ar-H), 7.95 (d, *J* = 9.0 Hz, 1H, Ar-H), 7.89 (dd, *J* = 9.0, 2.0 Hz, 1H, Ar-H), 7.57 (d, *J* = 4.5 Hz, 1H, Ar-H), 7.01 (t, *J* = 9.0 Hz, 2H, Ar-H), 6.84 – 6.75 (m, 2H, Ar-H), 6.15 (t, *J* = 6.5 Hz, 1H, NH), 4.31 (d, *J* = 6.5 Hz, 2H, CH_2). ESI-MS: m/z = 355 $[M+H]^+$.

*4.1.12.12. Tert-butyl 4-(3-(6-bromoquinolin-4-yl)prop-2-ynyl)piperazine-1-carboxylate (**14l**)*

This compound was prepared from 6-bromo-4-iodoquinoline (**12**) (400 mg, 1.20 mmol) and tert-butyl 4-(prop-2-ynyl)piperazine-1-carboxylate (**13l**) (269 mg, 1.20 mmol) according to the general synthesis procedure E to afford the title compound (224 mg, 0.52 mmol, 43% yield) as an off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.93 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.36 (d, *J* = 2.0 Hz, 1H, Ar-H), 8.03 (d, *J* = 9.0 Hz, 1H, Ar-H), 7.97 (dd, *J* = 9.0, 2.0 Hz, 1H, Ar-H), 7.70 (d, *J* = 4.5 Hz, 1H, Ar-H), 3.82 (s, 2H, CH_2), 3.40 (m, 4H, CH_2 × 2), 2.60 – 2.55 (m, 4H, CH_2 × 2), 1.40 (s, 9H, CH_3 × 3). ESI-MS: m/z = 430 $[M+H]^+$.

*4.1.13. General procedure F for suzuki coupling reaction (**15a-n**, **19a-e**)*

To a three-neck round bottom flask was added a halide (1.0 equiv), a boronic ester (1.0 equiv), $Pd(dppf)_2Cl_2$ (0.1 equiv) and K_2CO_3 (3.0 equiv) in dioxane/H_2O (3/1). The flask was fitted with a N_2 inlet adaptor and purged with N_2 for 15 min. The reaction mixture was then sealed under an atmosphere of N_2 and stirred at 100 °C for 10 h. The crude mixture was concentrated under reduced pressure and the residue was dissolved in CH_2Cl_2, washed with water twice, then the organic phase was dried over magnesium sulfate. The crude product was purified by silica gel column chromatography to give the desired target compound.

4.1.13.1.

*2,4-Difluoro-N-(5-(4-(4-hydroxybut-1-ynyl)quinolin-6-yl)-2-methoxypyridin-3-yl)benzenesulfonamide (**15a**)*

This compound was prepared from **14a** (33 mg, 0.12 mmol) and **5a** (50 mg, 0.12 mmol) according to the general synthesis procedure F to afford the title compound (25 mg, 0.051 mmol, 43% yield) as an off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.36 (s, 1H, NH), 8.85 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.48 (d, *J* = 2.5 Hz, 1H, Ar-H), 8.39 (d, *J* = 2.0 Hz, 1H, Ar-H), 8.12 (d, *J* = 9.0 Hz, 1H, Ar-H), 8.07 (dd, *J* = 9.0, 2.0 Hz, 1H, Ar-H), 8.03 (d, *J* = 2.5 Hz, 1H, Ar-H), 7.77 (m, 1H, Ar-H), 7.59 (d, *J* = 4.5 Hz, 1H, Ar-H), 7.56(m, 1H, Ar-H), 7.20 (m, 1H, Ar-H), 5.04 (t, *J* = 5.5 Hz, 1H, OH), 3.72 (q, *J* = 6.5 Hz, 2H, CH_2), 3.68 (s, 3H, OCH_3), 2.78 (t, *J* = 6.5 Hz, 2H, CH_2). ^{13}C NMR (100 MHz, DMSO-d_6) δ 165.06 (dd, $J_{C\text{-}F}$ = 252.8, 11.2 Hz), 159.36 (dd, $J_{C\text{-}F}$ = 255.9, 13.4 Hz), 157.59, 150.31, 146.88, 142.62, 135.02, 133.96, 131.83 (d, $J_{C\text{-}F}$ = 11.2 Hz), 130.45, 129.41, 128.75, 128.73, 127.53, 125.01 (dd, $J_{C\text{-}F}$ = 14.2, 3.3 Hz), 124.04, 122.42, 119.82, 111.85 (dd, $J_{C\text{-}F}$ = 22.8, 3.8 Hz), 105.81 (t, $J_{C\text{-}F}$ = 25.8 Hz), 100.20, 76.65, 59.48, 53.49, 23.69. ESI-MS: m/z = 496 $[M+H]^+$.

4.1.13.2.

*2,4-Difluoro-N-(2-methoxy-5-(4-(3-(piperidin-1-yl)prop-1-ynyl)quinolin-6-yl)pyridin-3-yl)benzenesulfonamide (**15b**)*

This compound was prepared from **14b** (39 mg, 0.12 mmol) and **5a** (50 mg, 0.12 mmol) according to the general synthesis procedure F to afford the title compound (16 mg, 0.029 mmol, 24% yield) as an off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.38 (s, 1H, NH), 8.87 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.42 (d, *J* = 2.5 Hz, 1H, Ar-H), 8.39 (d, *J* = 2.0 Hz, 1H, Ar-H), 8.14 (d, *J* = 9.0 Hz, 1H, Ar-H), 8.09 (dd, *J* = 9.0, 2.0 Hz, 1H, Ar-H), 7.99 (d, *J* = 2.5 Hz, 1H, Ar-H), 7.74 (m, 1H, Ar-H), 7.64 (d, *J* = 4.5 Hz, 1H, Ar-H), 7.53 (m, 1H, Ar-H), 7.17 (m, 1H, Ar-H), 3.74 (s, 2H, CH_2), 3.66 (s, 3H, OCH_3), 2.61 (m, 4H, CH_2 × 2), 1.59 – 1.52 (m, 4H, CH_2 × 2), 1.35 (m, 2H, CH_2). ^{13}C NMR (100 MHz, DMSO-d_6) δ 164.98 (dd, $J_{C\text{-}F}$ = 251.7, 11.0 Hz), 159.36 (dd, $J_{C\text{-}F}$ = 254.3, 11.8 Hz), 157.72, 150.35, 146.88, 142.41, 135.31, 133.76, 131.77 (d, $J_{C\text{-}F}$ =

10.0 Hz), 130.55, 128.82, 128.71, 127.28, 125.18 (d, J_{C-F} = 13.6 Hz), 124.21, 122.31, 120.27, 111.75 (dd, J_{C-F} = 21.9, 3.8 Hz), 105.73 (t, J_{C-F} = 25.6 Hz), 96.66, 80.48, 53.42, 52.63, 47.68, 25.37, 23.39. ESI-MS: m/z = 549 $[M+H]^+$.

4.1.13.3.

*2,4-Difluoro-N-(2-methoxy-5-(4-(3-(4-methylpiperazin-1-yl)prop-1-ynyl)quinolin-6-yl)pyridin-3-yl)benzenesulfonamide (**15c**)*

This compound was prepared from **14c** (41 mg, 0.12 mmol) and **5a** (50 mg, 0.12 mmol) according to the general synthesis procedure F to afford the title compound (21 mg, 0.037 mmol, 31% yield) as an off-white solid. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.85 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.36 (s, 1H, Ar-H), 8.26 (s, 1H, Ar-H), 8.17 (d, *J* = 9.0 Hz, 1H, Ar-H), 8.12 (s, 1H, Ar-H), 7.88 (m, 2H, Ar-H), 7.53 (d, *J* = 4.5 Hz, 1H, Ar-H), 7.00 – 6.90 (m, 2H, Ar-H), 3.95 (s, 3H, OCH_3), 3.75 (s, 2H, CH_2), 2.82 (s, 4H, CH_2 × 2), 2.62 (s, 4H, CH_2 × 2), 2.36 (s, 3H, CH_3). ^{13}C NMR (125 MHz, DMSO-d_6) δ 165.12 (dd, J_{C-F} = 251.0, 11.0 Hz), 159.81 (dd, J_{C-F} = 255.5, 13.5 Hz), 158.14, 150.67, 147.33, 141.13, 136.13, 132.53, 132.15 (d, J_{C-F} = 11.1 Hz), 130.98, 129.25, 129.02, 128.99, 127.78, 126.73 (dd, J_{C-F} = 15.6, 4.5 Hz), 124.75, 123.06, 122.50, 112.02 (dd, J_{C-F} = 21.6, 3.1 Hz), 106.08 (t, J_{C-F} = 26.1 Hz), 96.68, 81.08, 54.63, 53.79, 51.13, 47.24, 45.41. ESI-MS: m/z = 564 $[M+H]^+$.

4.1.13.4.

*2,4-Difluoro-N-(5-(4-(3-(4-hydroxypiperidin-1-yl)prop-1-ynyl)quinolin-6-yl)-2-metho xypyridin-3-yl)benzenesulfonamide (**15d**)*

This compound was prepared from **14d** (41 mg, 0.12 mmol) and **5a** (50 mg, 0.12 mmol) according to the general synthesis procedure F to afford the title compound (18 mg, 0.032 mmol, 27% yield) as an off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.36 (s, 1H, NH), 8.89 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.43 (d, *J* = 2.5 Hz, 1H, Ar-H), 8.38 (d, *J* = 2.0 Hz, 1H, Ar-H), 8.16 (d, *J* = 9.0 Hz, 1H, Ar-H), 8.11 (dd, *J* = 9.0, 2.0 Hz, 1H, Ar-H), 8.01 (d, *J* = 2.5 Hz, 1H, Ar-H), 7.78 (m, 1H, Ar-H), 7.67 (d, *J* = 4.5 Hz, 1H, Ar-H), 7.55 (m, 1H, Ar-H), 7.21 (m, 1H, Ar-H), 4.52 (s, 1H, OH), 3.76 (s, 2H,

CH_2), 3.70 (s, 3H, OCH_3), 3.45 (m, 1H, CH), 2.87 (m, 2H, CH_2), 2.42 (m, 2H, CH_2), 1.79 (m, 2H, CH_2), 1.46 (m, 2H, CH_2). ^{13}C NMR (100 MHz, DMSO-d_6) δ 164.98 (dd, J_{C-F} = 251.6, 9.6 Hz), 159.41 (dd, J_{C-F} = 267.8, 13.4 Hz), 157.68, 150.34, 146.86, 142.37, 135.28, 133.75, 131.77 (d, J_{C-F} = 10.3 Hz), 130.54, 128.81, 128.67, 128.64, 127.28, 125.16 (dd, J_{C-F} = 13.5, 4.2 Hz), 124.31, 122.27, 120.23, 111.81 (dd, J_{C-F} = 21.2, 3.8 Hz), 105.77 (t, J_{C-F} = 25.7 Hz), 96.63, 80.34, 63.7, 53.47, 49.75, 47.03, 34.19. ESI-MS: m/z = 565 $[M+H]^+$.

4.1.13.5.

*2,4-Difluoro-N-(5-(4-(3-(3-hydroxypiperidin-1-yl)prop-1-ynyl)quinolin-6-yl)-2-metho xypyridin-3-yl)benzenesulfonamide (**15e**)*

This compound was prepared from **14e** (41 mg, 0.12 mmol) and **5a** (50 mg, 0.12 mmol) according to the general synthesis procedure F to afford the title compound (15 mg, 0.027 mmol, 23% yield) as an off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.38 (s, 1H, NH), 8.90 (d, J = 4.5 Hz, 1H, Ar-H), 8.47 (d, J = 2.5 Hz, 1H, Ar-H), 8.40 (d, J = 2.0 Hz, 1H, Ar-H), 8.17 (d, J = 9.0 Hz, 1H, Ar-H), 8.12 (dd, J = 9.0, 2.0 Hz, 1H, Ar-H), 8.03 (d, J = 2.5 Hz, 1H, Ar-H), 7.78 (m, 1H, Ar-H), 7.68 (d, J = 4.5 Hz, 1H, Ar-H), 7.60 – 7.54 (m, 1H, Ar-H), 7.21 (td, J = 8.5, 2.0 Hz, 1H, Ar-H), 4.67 (s, 1H, OH), 3.78 (d, J = 4.5 Hz, 2H, CH_2), 3.69 (s, 3H, OCH_3), 3.59 – 3.53 (m, 1H, CH_2), 2.96 – 2.91 (dd, J = 10.5, 3.5 Hz, 1H, CH_2), 2.82 (m, 1H, CH_2), 2.32 (t, J = 9.5 Hz, 1H, CH_2), 2.14 (t, J = 9.5 Hz, 1H, CH_2), 1.76 (m, 2H, CH_2), 1.49 (m, 1H, CH_2), 1.08 (m, 1H, CH_2). ^{13}C NMR (125 MHz, DMSO-d_6) δ 165.51 (dd, J_{C-F} = 251.8, 11.5 Hz), 159.86 (dd, J_{C-F} = 255.8, 12.7 Hz), 158.18, 150.79, 147.37, 142.96, 135.78, 134.30, 132.30 (d, J_{C-F} = 10.8 Hz), 131.03, 129.29, 129.22, 129.15, 127.76, 125.65 (d, J_{C-F} = 11.3 Hz), 124.78, 122.78, 120.62, 112.28 (dd, J_{C-F} = 21.6, 2.8 Hz), 106.24 (t, J_{C-F} = 25.9 Hz), 96.92, 81.03, 66.41, 60.37, 53.93, 52.16, 47.68, 33.19, 23.34. ESI-MS: m/z = 565 $[M+H]^+$.

4.1.13.6.

2,4-Difluoro-N-(5-(4-(3-(2-hydroxyethoxy)prop-1-ynyl)quinolin-6-yl)-2-methoxypyridi

*n-3-yl)benzenesulfonamide (**15f**)*

This compound was prepared from **14f** (37 mg, 0.12 mmol) and **5a** (50 mg, 0.12 mmol) according to the general synthesis procedure F to afford the title compound (25 mg, 0.048 mmol, 40% yield) as an off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.36 (s, 1H, NH), 8.90 (d, J = 4.5 Hz, 1H, Ar-H), 8.47 (d, J = 2.5 Hz, 1H, Ar-H), 8.32 (d, J = 2.0 Hz, 1H, Ar-H), 8.15 (d, J = 9.0 Hz, 1H, Ar-H), 8.09 (dd, J = 9.0, 2.0 Hz, 1H, Ar-H), 8.01 (d, J = 2.5 Hz, 1H, Ar-H), 7.76 (m, 1H, Ar-H), 7.68 (d, J = 4.5 Hz, 1H, Ar-H), 7.59 – 7.53 (m, 1H, Ar-H), 7.20 (m, 1H, Ar-H), 4.69 (s, 1H, OH), 4.63 (s, 2H, CH_2), 3.67 (s, 3H, OCH_3), 3.66 – 3.62 (m, 2H, CH_2), 3.58 (m, 2H, CH_2). ^{13}C NMR (100 MHz, DMSO-d_6) δ 165.07 (dd, J_{C-F} = 254.2, 12.8 Hz), 159.36 (dd, J_{C-F} = 255.7, 12.9 Hz), 157.66, 150.32, 146.87, 142.72, 135.39, 134.05, 131.83 (d, J_{C-F} = 10.7 Hz), 130.55, 128.98, 128.70, 128.10, 127.09, 125.01 (d, J_{C-F} = 13.6 Hz), 124.49, 122.15, 119.86, 111.85 (dd, J_{C-F} = 21.8, 2.7 Hz), 105.80 (t, J_{C-F} = 27.0 Hz), 96.79, 80.86, 71.63, 60.07, 58.24, 53.48. ESI-MS: m/z = 526 $[M+H]^+$.

4.1.13.7.

*2,4-Difluoro-N-(5-(4-(3-((2-hydroxyethyl)(methyl)amino)prop-1-ynyl)quinolin-6-yl)-2-methoxypyridin-3-yl)benzenesulfonamide (**15g**)*

This compound was prepared from **14g** (38 mg, 0.12 mmol) and **5a** (50 mg, 0.12 mmol) according to the general synthesis procedure F to afford the title compound (28 mg, 0.052 mmol, 43% yield) as an off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.34 (s, 1H, NH), 8.88 (d, J = 4.5 Hz, 1H, Ar-H), 8.45 (d, J = 2.5 Hz, 1H, Ar-H), 8.38 (d, J = 2.0 Hz, 1H, Ar-H), 8.14 (d, J = 9.0 Hz, 1H, Ar-H), 8.09 (dd, J = 9.0, 2.0 Hz, 1H, Ar-H), 8.01 (d, J = 2.5 Hz, 1H, Ar-H), 7.75 (m, 1H, Ar-H), 7.66 (d, J = 4.5 Hz, 1H, Ar-H), 7.55 (m, 1H, Ar-H), 7.19 (m, 1H, Ar-H), 4.47 (s, 1H, OH), 3.83 (s, 2H, CH_2), 3.67 (s, 3H, OCH_3), 3.54 (t, J = 6.0 Hz, 2H, CH_2), 2.62 (t, J = 6.0 Hz, 2H, CH_2), 2.42 (s, 3H, CH_3). ^{13}C NMR (100 MHz, DMSO-d_6) δ 165.05 (dd, J_{C-F} = 251.7, 9.6 Hz), 159.52 (dd, J_{C-F} = 255.6, 8.6 Hz), 157.78, 150.40, 146.94, 142.45, 135.32, 133.91, 131.82 (d, J_{C-F} = 10.6 Hz), 130.61, 128.90, 128.76, 128.72, 127.32, 125.27 (dd, J_{C-F} = 13.8, 3.6 Hz), 124.45, 122.25, 120.30, 111.85 (dd, J_{C-F} = 22.0, 2.9 Hz),

105.82 (t, J_{C-F} = 25.3 Hz), 96.45, 80.60, 58.96, 57.78, 54.93, 46.43, 42.08. ESI-MS: m/z = 539 $[M+H]^+$.

4.1.13.8.

*N-(5-(4-(3-((2,3-Dihydroxypropyl)(methyl)amino)prop-1-ynyl)quinolin-6-yl)-2-methoxypyridin-3-yl)-2,4-difluorobenzenesulfonamide (**15h**)*

This compound was prepared from **14h** (42 mg, 0.12 mmol) and **5a** (50 mg, 0.12 mmol) according to the general synthesis procedure F to afford the title compound (33 mg, 0.058 mmol, 48% yield) as an off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.32 (s, 1H, NH), 8.88 (s, 1H, Ar-H), 8.44 (s, 1H, Ar-H), 8.38 (s, 1H, Ar-H), 8.14 (d, *J* = 8.5 Hz, 1H, Ar-H), 8.08 (d, *J* = 7.5 Hz, 1H, Ar-H), 8.00 (s, 1H, Ar-H), 7.75 (m, 1H, Ar-H), 7.66 (s, 1H, Ar-H), 7.55 (m, 1H, Ar-H), 7.19 (m, 1H, Ar-H), 4.48 (s, 2H, OH × 2), 3.84 (s, 2H, CH_2), 3.67 (s, 3H, OCH_3), 3.63 (s, 1H, CH), 3.35 (m, 2H, CH_2), 2.59 (m, 1H, CH_2), 2.41 (m, 4H, CH_3 + CH_2). ^{13}C NMR (125 MHz, DMSO-d_6) δ 165.49 (dd, J_{C-F} = 252.6, 11.6 Hz), 159.85 (dd, J_{C-F} = 256.0, 13.6 Hz), 158.19, 150.79, 147.39, 142.84, 135.78, 134.26, 132.27 (d, J_{C-F} = 10.6 Hz), 131.03, 129.33, 129.24, 129.19, 127.77, 125.73 (dd, J_{C-F} = 14.9, 4.0 Hz), 124.91, 122.73, 120.76, 112.28 (dd, J_{C-F} = 22.0, 2.9 Hz), 106.24 (t, J_{C-F} = 25.9 Hz), 96.96, 81.03, 69.90, 65.00, 59.34, 53.95, 47.26, 42.89. ESI-MS: m/z = 569 $[M+H]^+$.

4.1.13.9.

*2,4-Difluoro-N-(2-methoxy-5-(4-(3-(phenylamino)prop-1-ynyl)quinolin-6-yl)pyridin-3-yl)benzenesulfonamide (**15i**)*

This compound was prepared from **14i** (40 mg, 0.12 mmol) and **5a** (50 mg, 0.12 mmol) according to the general synthesis procedure F to afford the title compound (18 mg, 0.032 mmol, 27% yield) as an off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.39 (s, 1H, NH), 8.87 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.36 (d, *J* = 2.5 Hz, 1H, Ar-H), 8.25 (d, *J* = 2.0 Hz, 1H, Ar-H), 8.13 (d, *J* = 9.0 Hz, 1H, Ar-H), 8.05 (dd, *J* = 9.0, 2.0 Hz, 1H, Ar-H), 8.00 (d, *J* = 2.5 Hz, 1H, Ar-H), 7.78 (m, 1H, Ar-H), 7.60 (d, *J* = 4.5 Hz, 1H, Ar-H), 7.59 (m, 1H, Ar-H), 7.24 – 7.21 (m, 1H, Ar-H), 7.09 (m, 2H, Ar-H),

6.80 (m, 2H, Ar-H), 6.54 (t, *J* = 7.5 Hz, 1H, Ar-H), 6.21 (t, *J* = 6.5 Hz, 1H, NH), 4.37 (d, *J* = 6.5 Hz, 2H, CH_2), 3.73 (s, 3H, OCH_3). ^{13}C NMR (100 MHz, DMSO-d_6) δ 165.13 (d, J_{C-F} = 257.2 Hz), 159.46 (d, J_{C-F} = 255.2 Hz), 157.70, 150.29, 147.59, 146.82, 142.83, 142.71, 135.36, 134.24, 131.81 (d, J_{C-F} = 10.1 Hz), 130.53, 130.48, 129.01, 128.85, 128.75, 128.67, 127.33, 125.17 (d, J_{C-F} = 14.4 Hz), 124.21, 122.29, 119.96, 116.90, 112.99, 111.79 (dd, J_{C-F} = 21.7, 3.4 Hz), 105.78 (t, J_{C-F} = 25.4 Hz), 99.28, 77.18, 53.36, 32.97. ESI-MS: m/z = 557 $[M+H]^+$.

4.1.13.10.

*2,4-Difluoro-N-(2-methoxy-5-(4-(3-(4-methoxyphenylamino)prop-1-ynyl)quinolin-6-yl)pyridin-3-yl)benzenesulfonamide (**15j**)*

This compound was prepared from **14j** (44 mg, 0.12 mmol) and **5a** (50 mg, 0.12 mmol) according to the general synthesis procedure F to afford the title compound (22 mg, 0.038 mmol, 32% yield) as an off-white solid. ^1H NMR (500 MHz, DMSO-d_6) δ 10.42 (s, 1H, NH), 8.87 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.39 (d, *J* = 2.5 Hz, 1H, Ar-H), 8.27 (d, *J* = 2.0 Hz, 1H, Ar-H), 8.14 (d, *J* = 9.0 Hz, 1H, Ar-H), 8.06 (dd, *J* = 9.0, 2.0 Hz, 1H, Ar-H), 8.02 (d, *J* = 2.5 Hz, 1H, Ar-H), 7.79 (m, 1H, Ar-H), 7.59 (d, *J* = 4.5 Hz, 1H, Ar-H), 7.58 (m, 1H, Ar-H), 7.22 (m, 1H, Ar-H), 6.77 (d, *J* = 9.0 Hz, 2H, Ar-H), 6.71 (d, *J* = 9.0 Hz, 2H, Ar-H), 5.83 (t, *J* = 5.0 Hz, 1H, NH), 4.31 (d, *J* = 5.0 Hz, 2H, CH_2), 3.73 (s, 3H, OCH_3), 3.53 (s, 3H, OCH_3). ^{13}C NMR (125 MHz, DMSO-d_6) δ 165.51 (dd, J_{C-F} = 252.3, 11.8 Hz), 159.86 (dd, J_{C-F} = 256.3, 13.5 Hz), 158.13, 151.99, 150.75, 147.34, 142.87, 142.16, 135.83, 134.30, 132.29 (d, J_{C-F} = 10.4 Hz), 130.96, 129.41, 129.24, 129.22, 127.87, 125.77 (d, J_{C-F} = 13.8 Hz), 124.61, 123.44, 122.78, 114.97, 114.76, 112.28 (dd, J_{C-F} = 21.8, 3.1 Hz), 106.25 (t, J_{C-F} = 26.1 Hz), 100.18, 77.74, 55.56, 53.93, 34.45. ESI-MS: m/z = 587 $[M+H]^+$.

4.1.13.11.

*2,4-Difluoro-N-(5-(4-(3-(4-fluorophenylamino)prop-1-ynyl)quinolin-6-yl)-2-methoxyp yridin-3-yl)benzenesulfonamide (**15k**)*

This compound was prepared from **14k** (42 mg, 0.12 mmol) and **5a** (50 mg, 0.12 mmol) according to the general synthesis procedure F to afford the title compound (28 mg, 0.049 mmol, 41% yield) as an off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.41 (s, 1H, NH), 8.87 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.33 (d, *J* = 2.5 Hz, 1H, Ar-H), 8.24 (d, *J* = 2.0 Hz, 1H, Ar-H), 8.13 (d, *J* = 9.0 Hz, 1H, Ar-H), 8.05 (dd, *J* = 9.0, 2.0 Hz, 1H, Ar-H), 8.00 (d, *J* = 2.5 Hz, 1H, Ar-H), 7.78 (m, 1H, Ar-H), 7.60 (d, *J* = 4.5 Hz, 1H, Ar-H), 7.59 (m, 1H, Ar-H), 7.21 (m, 1H, Ar-H), 6.92 (t, *J* = 8.5 Hz, 2H, Ar-H), 6.83 – 6.76 (m, 2H, Ar-H), 6.19 (t, *J* = 6.5 Hz, 1H, NH), 4.35 (d, *J* = 6.5 Hz, 2H, CH_2), 3.73 (s, 3H, OCH_3). ^{13}C NMR (100MHz, DMSO-d_6) δ 165.08 (dd, J_{C-F} = 251.5, 12.4 Hz), 159.39 (dd, J_{C-F} = 255.3, 12.2 Hz), 157.68, 156.08, 150.29, 146.85, 144.23, 142.69, 135.30, 134.21, 131.82 (d, J_{C-F} = 10.8 Hz), 130.50, 129.34, 128.95, 128.68, 128.62, 128.23, 127.68, 127.31, 125.91, 125.61 (d, J_{C-F} = 12.3 Hz), 124.15, 122.22, 119.74, 115.39, 115.17, 113.92, 113.85, 111.83 (d, J_{C-F} = 23.8 Hz), 105.77 (t, J_{C-F} = 27.0 Hz), 99.17, 77.29, 53.42, 33.59. ESI-MS: m/z = 575 $[M+H]^+$.

4.1.13.12.

*2,4-Difluoro-N-(2-methoxy-5-(quinolin-6-yl)pyridin-3-yl)benzenesulfonamide (**15l**)*

This compound was prepared from **16** (25 mg, 0.12 mmol) and **5a** (50 mg, 0.12 mmol) according to the general synthesis procedure F to afford the title compound (23 mg, 0.054 mmol, 45% yield) as an off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.36 (s, 1H, NH), 8.93 (dd, *J* = 4.0, 1.5 Hz, 1H, Ar-H), 8.51 (d, *J* = 2.0 Hz, 1H, Ar-H), 8.45 (dd, *J* = 8.5, 1.0 Hz, 1H, Ar-H), 8.27 (d, *J* = 2.0 Hz, 1H, Ar-H), 8.12 (d, *J* = 9.0 Hz, 1H, Ar-H), 8.06 (m, 2H, Ar-H), 7.79 (m, 1H, Ar-H), 7.62 – 7.56 (m, 2H, Ar-H), 7.23 (m, 1H, Ar-H), 3.68 (s, 3H, OCH_3). ^{13}C NMR (125 MHz, DMSO-d_6) δ 165.56 (dd, J_{C-F} = 252.5, 11.8 Hz), 159.88 (dd, J_{C-F} = 256.1, 13.4 Hz), 158.13, 151.23, 147.58, 143.16, 136.73, 134.78, 134.57, 132.36 (d, J_{C-F} = 16.0 Hz), 130.24, 129.37, 128.65, 128.56, 126.65, 125.56 (dd, J_{C-F} = 13.8, 3.6 Hz), 122.46, 120.22, 112.31 (dd, J_{C-F} = 22.1, 3.1 Hz), 106.27 (t, J_{C-F} = 25.8 Hz), 53.90. ESI-MS: m/z = 428 $[M+H]^+$.

*4.1.14. 6-Bromo-4-(3-(piperazin-1-yl)prop-1-ynyl)quinoline (**17**)*

A solution of tert-butyl 4-(3-(6-bromoquinolin-4-yl)prop-2-ynyl)piperazine-1-carboxylate (**14l**) (200 mg, 0.47 mmol) in CH_2Cl_2 (10 mL) was treated with TFA (10 mL) and stirred at room temperature for 1 h. The crude mixture was concentrated under reduced pressure and the residue was dissolved in CH_2Cl_2, washed with 1 N NaOH and water. The organic phase was dried over magnesium sulfate to give the title compound (130 mg, 0.40 mmol, 85% yield) as a viscous oil. ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.91 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.33 (d, *J* = 2.0 Hz, 1H, Ar-H), 7.99 (t, *J* = 9.0 Hz, 1H, Ar-H), 7.94 (dd, *J* = 9.0, 2.0 Hz, 1H, Ar-H), 7.65 (t, *J* = 4.5 Hz, 1H, Ar-H), 3.71 (s, 2H, CH_2), 2.77 (s, 4H, CH_2 × 2), 2.54 (s, 4H, CH_2 × 2). ESI-MS: m/z = 330 $[M+H]^+$.

*4.1.15.1 1-(4-(3-(6-Bromoquinolin-4-yl)prop-2-ynyl)piperazin-1-yl)ethanone (**18a**)*

A solution of 6-bromo-4-(3-(piperazin-1-yl)prop-1-ynyl)quinoline (**17**) (60 mg, 0.18 mmol) was dissolved in anhydrous CH_2Cl_2 (10 mL) followed by addition of anhydrous Et_3N (75 µl, 0.54 mmol). A solution of acetyl chloride (13 µl, 0.18 mmol) in anhydrous CH_2Cl_2 (2 mL) was then syringed into the reaction mixture slowly and the solution was stirred for 10 min at 0 °C. The reaction mixture was washed with 1 N NaOH, water and brine. The organic layer was dried over magnesium sulfate to give title compound as a white solid (56 mg, 0.15 mmol, 83% yield). ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.93 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.36 (d, *J* = 2.0 Hz, 1H, Ar-H), 8.03 (d, *J* = 9.0 Hz, 1H, Ar-H), 7.97 (dd, *J* = 9.0, 2.0 Hz, 1H, Ar-H), 7.70 (d, *J* = 4.5 Hz, 1H, Ar-H), 3.84 (s, 2H, CH_2), 3.56 – 3.48 (m, 4H, CH_2 × 2), 2.68 – 2.60 (m, 2H, CH_2), 2.59 – 2.55 (m, 2H, CH_2), 2.02 (s, 3H, CH_3). ESI-MS: m/z = 372 $[M+H]^+$.

*4.1.15.2. 6-Bromo-4-(3-(4-(methylsulfonyl)piperazin-1-yl)prop-1-ynyl)quinoline (**18b**)*

This compound was prepared from 6-bromo-4-(3-(piperazin-1-yl)prop-1-ynyl)quinoline (**17**) (60 mg, 0.18 mmol), methanesulfonyl chloride (14 µl, 0.18 mmol) and anhydrous Et_3N (75 µl, 0.54 mmol) according to the above synthesis procedure to afford the title compound as a white solid (65 mg, 0.16 mmol, 89% yield). ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.94 (d, *J* =

4.5 Hz, 1H, Ar-H), 8.36 (d, *J* = 2.0 Hz, 1H, Ar-H), 8.03 (d, *J* = 9.0 Hz, 1H, Ar-H), 7.98 (dd, *J* = 9.0, 2.0 Hz, 1H, Ar-H), 7.72 (d, *J* = 4.5 Hz, 1H, Ar-H), 3.86 (s, 2H, CH_2), 3.20 (m, 4H, CH_2 × 2), 2.91 (s, 3H, CH_3), 2.73 (m, 4H, CH_2 × 2). ESI-MS: m/z = 408 $[M+H]^+$.

4.1.16.1

*N-(5-(4-(3-(4-Acetylpiperazin-1-yl)prop-1-ynyl)quinolin-6-yl)-2-methoxypyridin-3-yl) -2,4-difluorobenzenesulfonamide (**15m**)*

This compound was prepared from **18a** (45 mg, 0.12 mmol) and **5a** (50 mg, 0.12 mmol) according to the general synthesis procedure F to afford the title compound (32 mg, 0.054 mmol, 45% yield) as an off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.41 (s, 1H, NH), 8.91 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.48 (d, *J* = 2.0 Hz, 1H, Ar-H), 8.36 (d, *J* = 2.0 Hz, 1H, Ar-H), 8.17 (d, *J* = 9.0 Hz, 1H, Ar-H), 8.13 (dd, *J* = 9.0, 2.0 Hz, 1H, Ar-H), 8.04 (d, *J* = 2.0 Hz, 1H, Ar-H), 7.79 (m, 1H, Ar-H), 7.69 (d, *J* = 4.5 Hz, 1H, Ar-H), 7.62 – 7.56 (m, 1H, Ar-H), 7.22 (m, 1H, Ar-H), 3.84 (s, 2H, CH_2), 3.69 (s, 3H, OCH_3), 3.51 (m, 4H, CH_2 × 2), 2.68 (t, *J* = 5.0 Hz, 2H, CH_2), 2.58 (t, *J* = 5.0 Hz, 2H, CH_2), 1.96 (s, 3H, CH_3). ^{13}C NMR (125 MHz, DMSO-d_6) δ 168.69, 165.55 (dd, J_{C-F} = 252.9, 11.0 Hz), 159.87 (dd, J_{C-F} = 256.1, 13.0 Hz), 158.17, 150.81, 147.38, 142.98, 135.75, 134.35, 132.29 (d, J_{C-F} = 11.0 Hz), 131.04, 129.31, 129.18, 128.99, 127.75, 125.60 (dd, J_{C-F} = 14.8, 4.1 Hz), 124.90, 122.68, 120.55, 112.32 (dd, J_{C-F} = 21.6, 2.9 Hz), 106.27 (t, J_{C-F} = 25.4 Hz), 96.55, 80.97, 53.97, 52.19, 51.74, 47.37, 46.00, 41.16. 21.56. ESI-MS: m/z = 592 $[M+H]^+$.

4.1.16.2.

*2,4-Difluoro-N-(2-methoxy-5-(4-(3-(4-(methylsulfonyl)piperazin-1-yl)prop-1-ynyl)qui nolin-6-yl)pyridin-3-yl)benzenesulfonamide (**15n**)*

This compound was prepared from **18b** (49 mg, 0.12 mmol) and **5a** (50 mg, 0.12 mmol) according to the general synthesis procedure F to afford the title compound (42 mg, 0.067 mmol, 56% yield) as a off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.39 (s, 1H, NH), 8.91 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.44 (s, 1H, Ar-H), 8.36 (d, *J* = 2.0

Hz, 1H, Ar-H), 8.17 (d, *J* = 9.0 Hz, 1H, Ar-H), 8.12 (dd, *J* = 9.0, 2.0 Hz, 1H, Ar-H), 8.03 (d, *J* = 2.0 Hz, 1H, Ar-H), 7.79 (m, 1H, Ar-H), 7.70 (d, *J* = 4.5 Hz, 1H, Ar-H), 7.56 (t, *J* = 9.0 Hz, 1H, Ar-H), 7.21 (m, 1H, Ar-H), 3.86 (s, 2H, CH_2), 3.70 (s, 3H, OCH_3), 3.20 (m, 4H, CH_2 × 2), 2.88 (s, 3H, CH_3), 2.75 (m, 4H, CH_2 × 2). ^{13}C NMR (100 MHz, DMSO-d_6) δ 164.97 (dd, J_{C-F} = 253.7, 11.0 Hz), 159.50 (dd, J_{C-F} = 256.2, 11.1 Hz), 157.65, 150.35, 146.90, 142.56, 135.30, 133.88, 131.81 (d, J_{C-F} = 10.1 Hz), 130.57, 128.86, 128.68, 128.46, 127.25, 125.06 (dd, J_{C-F} = 12.0, 4.3 Hz), 124.47, 122.20, 120.04, 111.84 (dd, J_{C-F} = 21.9, 3.0 Hz), 105.79 (t, J_{C-F} = 26.7 Hz), 96.03, 80.43, 53.47, 50.86, 46.66, 45.28, 33.87. ESI-MS: m/z = 628 $[M+H]^+$.

4.1.17.1.

*4-Fluoro-N-(5-(4-(3-(4-hydroxypiperidin-1-yl)prop-1-ynyl)quinolin-6-yl)-2-methoxyp yridin-3-yl)benzenesulfonamide (**19a**)*

This compound was prepared from **14d** (41 mg, 0.12 mmol) and **5b** (49 mg, 0.12 mmol) according to the general synthesis procedure F to afford the title compound (32 mg, 0.059 mmol, 49% yield) as an off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.13 (s, 1H, NH), 8.90 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.43 (s, 1H, Ar-H), 8.39 (s, 1H, Ar-H), 8.17 (d, *J* = 8.5 Hz, 1H, Ar-H), 8.11 (d, *J* = 8.5 Hz, 1H, Ar-H), 8.02 (s, 1H, Ar-H), 7.83 (dd, *J* = 8.0, 5.0 Hz, 2H, Ar-H), 7.68 (d, *J* = 4.5 Hz, 1H, Ar-H), 7.41 (t, *J* = 8.5 Hz, 2H, Ar-H), 4.51 (d, *J* = 5.0 Hz, 1H, OH), 3.76 (s, 2H, CH_2), 3.70 (s, 3H, OCH_3), 3.45 (m, 1H, CH), 2.87 (m, 2H, CH_2), 2.42 (m, 2H, CH_2), 1.79 (m, 2H, CH_2), 1.47 (m, 2H, CH_2). ^{13}C NMR (125 MHz, DMSO-d_6) δ 164.80 (d, J_{C-F} = 249.9 Hz), 157.32, 150.80, 147.37, 142.13, 137.21 (d, J_{C-F} = 2.0 Hz), 135.86, 132.28, 131.04, 130.18 (d, J_{C-F} = 9.5 Hz), 129.24, 129.18, 129.11, 127.80, 124.80, 122.74, 121.29, 116.66 (d, J_{C-F} = 22.8 Hz), 97.28, 80.75, 66.31, 53.99, 50.30, 47.58, 34.77. ESI-MS: m/z = 547 $[M+H]^+$.

4.1.17.2.

*3-Fluoro-N-(5-(4-(3-(4-hydroxypiperidin-1-yl)prop-1-ynyl)quinolin-6-yl)-2-methoxyp yridin-3-yl)benzenesulfonamide (**19b**)*

This compound was prepared from **14d** (41 mg, 0.12 mmol) and **5c** (49 mg, 0.12 mmol) according to the general synthesis procedure F to afford the title compound (38 mg, 0.070 mmol, 58% yield) as an off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 10.25 (s, 1H, NH), 8.90 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.44 (d, *J* = 2.0 Hz, 1H, Ar-H), 8.38 (d, *J* = 1.5 Hz, 1H, Ar-H), 8.17 (d, *J* = 8.5 Hz, 1H, Ar-H), 8.10 (dd, *J* = 8.5, 1.5 Hz, 1H, Ar-H), 8.02 (t, *J* = 2.0 Hz, 1H, Ar-H), 7.68 (d, *J* = 4.5 Hz, 1H, Ar-H), 7.64 (m, 1H, Ar-H), 7.61 – 7.57 (m, 2H, Ar-H), 7.53 (m, 1H, Ar-H), 4.50 (s, 1H, OH), 3.77 (s, 2H, CH_2), 3.72 (s, 3H, OCH_3), 3.46 (m, 1H, CH), 2.90 – 2.84 (m, 2H, CH_2), 2.42 (m, 2H, CH_2), 1.79 (m, 2H, CH_2), 1.46 (m, 2H, CH_2). ^{13}C NMR (125 MHz, DMSO-d_6) δ 162.06 (d, $J_{C\text{-}F}$ = 247.1 Hz), 157.39, 150.81, 147.37, 142.98, 142.62 (d, $J_{C\text{-}F}$ = 6.6 Hz), 135.82, 132.51, 131.90 (d, $J_{C\text{-}F}$ =7.8 Hz), 131.03, 129.25, 129.16, 129.14, 127.79, 124.81, 123.36 (d, $J_{C\text{-}F}$ = 2.9 Hz), 122.75, 121.10, 120.48 (d, $J_{C\text{-}F}$ = 21.1 Hz), 114.07 (d, $J_{C\text{-}F}$ = 24.4 Hz), 97.19, 80.77, 66.22, 53.99, 50.27, 47.57, 34.72. ESI-MS: m/z = 547 $[M+H]^+$.

4.1.17.3.

*N-(5-(4-(3-(4-Hydroxypiperidin-1-yl)prop-1-ynyl)quinolin-6-yl)-2-methoxypyridin-3-yl)-4-(trifluoromethyl)benzenesulfonamide (**19c**)*

This compound was prepared from **14d** (41 mg, 0.12 mmol) and **5d** (55 mg, 0.12 mmol) according to the general synthesis procedure F to afford the title compound (31 mg, 0.052 mmol, 43% yield) as an off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.89 (t, *J* = 4.5 Hz, 1H, Ar-H), 8.45 (d, *J* = 2.5 Hz, 1H, Ar-H), 8.40 (d, *J* = 2.0 Hz, 1H, Ar-H), 8.16 (d, *J* = 9.0 Hz, 1H, Ar-H), 8.09 (dd, *J* = 9.0, 2.0 Hz, 1H, Ar-H), 8.04 (d, *J* = 2.5 Hz, 1H, Ar-H), 8.01 – 7.93 (m, 4H, Ar-H), 7.67 (d, *J* = 4.5 Hz, 1H, Ar-H), 4.55 (s, 1H, OH), 3.77 (s, 2H, CH_2), 3.61 (s, 3H, OCH_3), 3.46 (m, 1H, CH), 2.88 (m, 2H, CH_2), 2.44 (m, 2H, CH_2), 1.80 (m, 2H, CH_2), 1.46 (m, 2H, CH_2). ^{13}C NMR (125 MHz, DMSO-d_6) δ 157.67, 150.82, 147.38, 144.87, 142.57, 135.80, 133.34, 132.90 (d, $J_{C\text{-}F}$ = 32.3 Hz), 131.02, 129.28, 129.22, 129.18, 128.08, 127.80, 126.69 (d, $J_{C\text{-}F}$ = 3.6 Hz), 123.94 (d, $J_{C\text{-}F}$ = 271.0 Hz), 124.80, 122.79, 121.03, 97.20, 80.80, 66.24, 53.80, 50.26, 47.56, 34.71. ESI-MS: m/z = 597 $[M+H]^+$.

4.1.17.4.

*N-(5-(4-(3-(4-Hydroxypiperidin-1-yl)prop-1-ynyl)quinolin-6-yl)-2-methoxypyridin-3-yl)-4-(trifluoromethoxy)benzenesulfonamide (**19d**)*

This compound was prepared from **14d** (41 mg, 0.12 mmol) and **5e** (57 mg, 0.12 mmol) according to the general synthesis procedure F to afford the title compound (35 mg, 0.057 mmol, 48% yield) as an off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.90 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.46 (d, *J* = 2.5 Hz, 1H, Ar-H), 8.40 (d, *J* = 2.0 Hz, 1H, Ar-H), 8.16 (d, *J* = 9.0 Hz, 1H, Ar-H), 8.10 (dd, *J* = 9.0, 2.0 Hz, 1H, Ar-H), 8.03 (d, *J* = 2.5 Hz, 1H, Ar-H), 7.89 – 7.86 (d, *J* = 9.0 Hz, 2H, Ar-H), 7.67 (d, *J* = 4.5 Hz, 1H, Ar-H), 7.58 (d, *J* = 8.5 Hz, 2H, Ar-H), 4.54 (s, 1H, OH), 3.77 (s, 2H, CH_2), 3.62 (s, 3H, OCH_3), 3.44 (m, 1H, CH), 2.88 (m, 2H, CH_2), 2.44 (m, 2H, CH_2), 1.80 (m, 2H, CH_2), 1.46 (m, 2H, CH_2). ^{13}C NMR (125 MHz, DMSO-d_6) δ 157.67, 151.50, 150.83, 147.38, 142.69, 139.78, 135.79, 133.46, 131.02, 129.74, 129.27, 129.19, 127.80, 124.80, 122.79, 121.80, 121.34, 120.90, 119.29, 97.20, 80.80, 66.23, 53.80, 50.26, 47.56, 34.70. ESI-MS: m/z = 613 $[M+H]^+$.

4.1.17.5.

*N-(5-(4-(3-(4-Hydroxypiperidin-1-yl)prop-1-ynyl)quinolin-6-yl)-2-methoxypyridin-3-yl)-4-methylbenzenesulfonamide (**19e**)*

This compound was prepared from **14d** (41 mg, 0.12 mmol) and **5f** (48 mg, 0.12 mmol) according to the general synthesis procedure F to afford the title compound (28 mg, 0.052 mmol, 43% yield) as an off-white solid. ^{1}H NMR (500 MHz, DMSO-d_6) δ 9.98 (s, 1H, NH), 8.89 (d, *J* = 4.5 Hz, 1H, Ar-H), 8.39 (d, *J* = 2.0 Hz, 1H, Ar-H), 8.36 (d, *J* = 1.5 Hz, 1H, Ar-H), 8.15 (d, *J* = 8.5 Hz, 1H, Ar-H), 8.05 (dd, *J* = 8.5, 1.5 Hz, 1H, Ar-H), 8.00 (d, *J* = 2.0 Hz, 1H, Ar-H), 7.68 (d, *J* = 8.0 Hz, 2H, Ar-H), 7.66 (d, *J* = 4.5 Hz, 1H, Ar-H), 7.36 (d, *J* = 8.0 Hz, 2H, Ar-H), 4.52 (s, 1H, OH), 3.76 (s, 2H, CH_2), 3.72 (s, 3H, OCH_3), 3.46 (m, 1H, CH), 2.87 (m, 2H, CH_2), 2.42 (m, 2H, CH_2), 2.35 (s, 3H, CH_3), 1.79 (m, 2H, CH_2), 1.46 (m, 2H, CH_2). ^{13}C NMR (125 MHz, DMSO-d_6) δ 156.89, 150.79, 147.36, 143.79, 141.54, 137.79, 135.92, 131.05, 130.88,

129.94, 129.17, 128.95, 127.80, 127.16, 124.81, 122.67, 121.65, 97.28, 80.72, 66.32, 54.02, 50.31, 47.59, 34.78, 21.41. ESI-MS: m/z = 543 $[M+H]^+$.

5. Biological assay

5.1. Class I PI3Ks enzyme assay

The inhibition of PI3Ks (P110α/85α, Promege; P110β, Millipore; P110γ, Invitrogen; P110δ, Millipore) activity was determined using the Kinase-Glo Plus Luminescent Kinase assay (PI3Kα, Promege) and ADP-Glo Kinase assay (PI3Kβ, γ and δ, Promege), respectively. Test compounds were serially diluted to the desired concentrations and then 2.5 μL of each of them was added to a 384-well plate (Corning) as assay plate. 1x kinase buffer was prepared contained 50 mM HEPES (pH 7.5), 3 mM $MgCl_2$, 1 mM EGTA, 100 mM NaCl, 0.03% CHAPS, 2 mM DTT. PI3K enzyme was diluted in the 1x kinase buffer to give 4x kinase solutions. PI3Kα, β, γ and δ were diluted to the final concentrations of 1.65 nM, 4.8 nM, 7.6 nM and 5.7 nM, respectively. 2.5 μL of kinase solution was then added to each well of the assay plate, except for control well without enzyme (add 2.5 μL of 1x kinase buffer instead). Meanwhile, PIP2 substrate and ATP were diluted in the 1x kinase buffer to give 2x substrate solution with final concentrations of 50 μM of PIP2 and 25 μM of ATP. After that, 5 μL of substrate solution was added to each well of the assay plate to start reaction. The assay plate was covered and incubated at room temperature for 1 h. As for PI3Kα, 10 μL of Kinase-Glo reagent was then added to each well of the assay plate to stop the reaction. Subsequently, the mixture was treated briefly with centrifuge, shaked slowly on the shaker for 15 min before reading on a plate reader for luminescence. As for PI3Kβ, γ and δ, 5 μL reaction mixture was transferred from 384-well to a new 384 plate and 5 μL of ADP-Glo reagent was added to each well to stop the reaction. The mixture was treated briefly with centrifuge, shaked slowly on the shaker and equilibrated for 40 min. 10 μL Kinase Detection reagent was added to each wells, shaked for 1 min and equilibrated for 1 h before reading on a plate reader for luminescence. Finally, conversion data was collected on Flex station and RLU values were converted to inhibition values using the formula of (sample RLU

-min)/(max-min) ×100. Herein, “min” means the RLU of no enzyme control and “max” means the RLU of DMSO control.

5.2. mTOR enzyme assay

The inhibition of mTOR (1362-end, Millipore) activity was determined by using the Lance Ultra assay. Test compounds were serially diluted to the desired concentrations and then 2.5 μL of each of them was added to a 384-well plate (Corning) as assay plate. 1x kinase buffer was prepared contained 50 mM HEPES (pH 7.5), 10 mM $MgCl_2$, 1 mM EGTA, 3 mM $MnCl_2$, 0.01% Tween-20, 2 mM DTT. mTOR was diluted in the 1x kinase buffer to give 4x kinase solution with a final concentration of 2.5 nM. 2.5 μL of kinase solution was then added to each well of the assay plate, except for control well without enzyme. Meanwhile, ULight-4E-BP1 peptide substrate (Thr37/46, PE) and ATP were diluted in the 1x kinase buffer to give 2x substrate solution with final concentrations of 50 μM of ULight-4E-BP1 peptide and 10.8 μM of ATP. 5 μL of substrate solution was then added to each well of the assay plate to start reaction. The assay plate was coved and incubated at room temperature for 1 hour. Subsequently, 10 μL of prepared detection solution buffer containing EDTA and Eu-anti-phospho-4E-BP1 antibody (Thr37/46, PE) was added to each well of the assay plate. After treating briefly with centrifuge, allow the plate to equilibrate for 60 minutes before reading on a plate reader. Finally, conversion data was collected on Envision and Lance signal (665nm) values were converted to inhibition values using the formula of (Lance signal-min)/(max-min) × 100. Herein, “min” means the Lance signal of no enzyme control and “max” means the Lance signal of DMSO control.

5.3. KinomeScan binding assay

Kinase selectivity of test compound was screened at the concentration of 10 μM by the KinomeScan binding assay (DiscoveRx), and the protocol of this assay is consistent with literature [27].

5.4. Anti-proliferative assay

All new synthesized 4-alkynyl-quinoline derivatives were screened for their anti-proliferative activity by SRB assay. PC-3 and HCT-116 cells were seeded into 96-well plates and cultured for 10 h. Subsequently, it was exposed to serial concentrations of compound for 72 h. Cells were then washed with PBS and fixed with 10% (w/v) trichloroacetic acid at 4 °C for 1 hour. After washing, the cells were stained for 30 min with 0.4% SRB dissolved in 1% acetic acid and then washed by 1% acetic acid for 5 times. Finally, the protein-bound dye was extracted using 10 mM unbuffered Tris base. The absorbance was obtained at 515 nm on a multiscan spectrum (Thermo Fisher). The inhibition rate on cell proliferation of each well was calculated using the formula of (A515 control cells - A515 treated cells)/A515 control cells × 100%.

5.5. Western blot assay

The suppressive activities of Akt and pAkt(Ser473) in PC-3 cells were determined by western blot analysis. GAPDH was employed as the internal control. PC-3 cells were seeded into six-well plate at 8×10^5 cells per well and then incubated at 37 °C overnight prior to drug exposure. Cells were treated with **15d** at various concentrations of 1 nM, 5 nM, 25 nM and 125 nM, and incubated at 37 °C for 3 h. After compound treatment, culture medium was discarded and cells were rinsed with pre-chilled PBS 3 times immediately. Subsequently, cell lysates were collected by adding RIPA lysis buffer (50 mM Tris-HCl pH 8.0, 150 mM NaCl, 1% NP-40, 0.5% sodium deoxycholate, 0.1% SDS) with protease and phosphatase inhibitor (Roche) onto cells. The cell lysates were clarified by centrifugation at 12,000 rpm for 15 min at 4 °C and the supernatant was collected as protein sample. The total protein concentration in protein sample was quantified using BCA protein Assay Kit (Pierce). Following this, the normalized protein samples were mixed with 4x NuPAGE LDS sample buffer (Life Tech) and boiled at 95˚C for 5 minute, then loaded and electrophoresed in NuPAGE 4-12 % Novex Bis-Tris gel (Life Tech). Subsequently, using Life Technologies IBLOT transfer system (Life Tech, IB301002), proteins were

transferred from gel to a nitrocellulose membrane, which was blocked in SuperBlock blocking buffer (Thermo Fisher) for 1 h then in primary antibody dilution (rabbit anti-pAkt(Ser473), rabbit anti-Akt, mouse anti-GAPDH, Abcam) overnight at 4 °C. After washing in TBST buffer (25 mM Tris-HCl pH 7.6, 150 mM NaCl, 0.1% Tween-20), membrane was incubated in diluted secondary antibodies for 2 h at room temperature, and washed in TBST again. Finally, membrane was imaged using LICOR Odyssey system.

6. Molecular docking study

The co-crystal structures of PI3Kγ with GSK2126458 (PDB code 3L08) and PI3Kα with BYL719 (PDB code 4JPS) were chosen as the templates to generate the docking modes. For the preparation of ligands, the 3D structures were generated and their energy minimization was performed by using Discovery Studio 2.1. For the preparation of protein, the hydrogen atoms were added, and the CHARMm-force field was employed. The whole enzyme was defined as a receptor and the site sphere was selected based on the ligand binding sites, then the ligand was removed and compound was placed during the molecular docking procedure. Types of interactions of the docked PI3K protein with compound were analyzed and then the docking conformations were selected and saved based on calculated energy.

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Figure captions：

Fig. 1. Structures of clinical PI3K/mTOR dual inhibitors.

Fig. 2. The design concept based on the co-crystal structure of GSK2126458 with PI3Kγ.

Fig. 3. The suppressive effect of **15d** on pAkt(Ser473) in PC-3 cells.

Fig. 4. Docking modes of compounds **15d** with PI3Kγ, and **15d**, **19a** and **19b** with PI3Kα, respectively. The hydrogen bonds are shown by dashed lines. (A) ribbon show of compound **15d** bound to PI3Kγ; (B, C and D) ribbon show of compounds **15d**, **19a** and **19b** bound to PI3Kα, respectively.

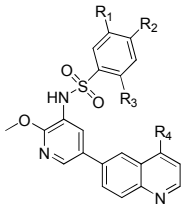
Scheme 1. The synthetic route for intermediates **5a-f**. Reagents and conditions: (a) $NaOCH_3$, CH_3OH, 0-50 °C, 12 h; (b) $SnCl_2$, EtOAc, 50 °C, 3 h; (c) benzenesulfonyl chloride, pyridine, rt, 24 h; (d) bis(pinacolato)diborane, $Pd(dppf)_2Cl_2$, AcOK, dioxane, 100 °C, 3 h.

Scheme 2. The synthetic route for target compounds **15a-l**. Reagents and conditions: (a) diethyl-2-(ethoxymethylene)malonate, 80 °C, 2 h; (b) Ph_2O, 250 °C, 6 h; (c) NaOH, 100 °C, 1 h, then HCl, rt, 10min; (d) Ph_2O, 260 °C, 2 h; (e) $POCl_3$, 120 °C, 6 h; (f) HCl/EA, rt, 30min, then KI, MeCN, reflux, 48 h; (g) $Pd(PPh_3)_2Cl_2$, CuI, Et_3N, rt, 5 h; (h) $Pd(dppf)_2Cl_2$, K_2CO_3, dioxane/H_2O, 100 °C, 10 h.

Scheme 3. The synthetic route for target compounds **15m**, **15n** and **19a-e**. Reagents and conditions: A. (a) TFA, CH_2Cl_2, rt, 1-2 h; (b) acetic chloride or methanesulfonyl chloride, CH_2Cl_2, Et_3N, 0 °C, 10 min; (c) $Pd(dppf)_2Cl_2$, K_2CO_3, dioxane/H_2O, 100 °C, 15 h; B. (a) $Pd(dppf)_2Cl_2$, K_2CO_3, dioxane/H_2O, 100 °C, 15 h.

Table 1

PI3Kα inhibitory activities and anti-proliferative activities of compounds **15a-n** and **19a-e** as well as their molecular parameters.



Compd.	R_4	R_1	R_2	R_3	PI3Kα IC_{50} (nM)	IC_{50} (μM)[a] PC-3	IC_{50} (μM)[a] HCT116	ClogP[b]	tPSA[b]
15l	H	H	F	F	14.28	7.16	3.67	4.71	80.12
15a		H	F	F	4.62	2.30	1.26	4.05	100.35
15b		H	F	F	27.33	1.64	1.60	5.66	83.36
15c		H	F	F	24.15	1.95	1.54	3.52	86.60
15d		H	F	F	1.63	0.37	2.47	3.58	103.59
15e		H	F	F	2.57	1.15	1.42	4.46	103.59
15f		H	F	F	2.34	1.10	1.02	3.54	109.58
15g		H	F	F	2.71	1.12	0.48	3.88	103.59
15h		H	F	F	2.21	0.66	0.49	3.86	123.82
15i		H	F	F	27.43	1.37	1.71	5.92	92.15
15j		H	F	F	32.54	7.76	6.23	6.03	101.38
15k		H	F	F	21.91	4.83	3.53	6.37	92.15
15m		H	F	F	2.02	3.25	1.21	4.03	103.67
15n		H	F	F	3.45	2.42	0.61	4.45	120.74
19a		H	F	H	39.76	1.40	2.14	3.41	103.59
19b		F	H	H	43.11	1.93	0.88	3.41	103.59
19c		H	CF_3	H	86.64	4.93	4.24	4.23	103.59
19d		H	OCF_3	H	> 300	6.21	6.26	4.42	112.82
19e		H	CH_3	H	151.23	2.15	3.37	3.68	103.59
BEZ-235					35.15	0.51	0.22	5.81	72.06
GSK2126458								4.07	104.84

[a]Values are means of three experiments

[b] Calculated by ChemBioDraw Ultra 11.0.

Table 2

Enzymatic activities of compound **15d** against PI3Ks and mTOR (IC_{50}, nM)[a]

Enzyme	Compd.	
	15d	BEZ235
PI3Kα	1.63	35.15
PI3Kβ	6.91	16.23
PI3Kγ	0.38	25.53
PI3Kδ	2.14	89.06
mTOR	3.26	20.74

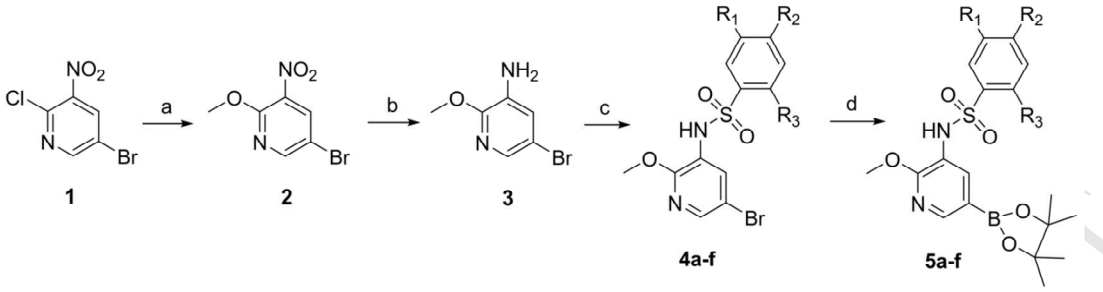
[a]Values are means of three experiments

Table 3 Kinase selectivity of **15d** in a panel of kinases (KinomeScan)[a]

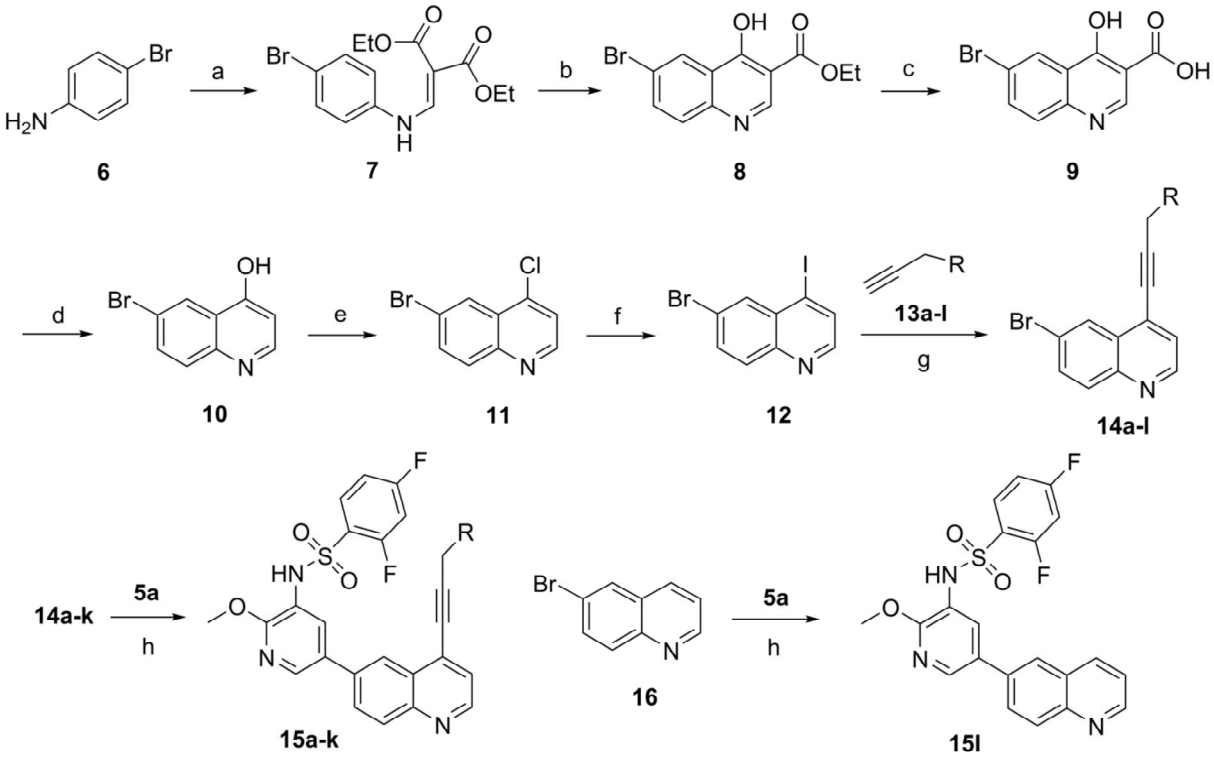
Kinase[b]	% Ctrl	Kinase	% Ctrl	Kinase	% Ctrl	Kinase	% Ctrl	Kinase	% Ctrl
AAK1	24	CDKL3	93	IRAK3	11	PCTK3	100	PIP5K1C	20
ACVR2A	100	CDKL5	100	LZK	76	PFTK1	100	PIP5K2B	100
ACVR2B	100	CIT	74	MAP3K15	57	PIK3C2B	0	PIP5K2C	7.5
ANKK1	25	CSNK1A1L	85	MAP3K4	92	PIK3C2G	0	PKMYT1	87
BIKE	12	EPHB6	7.6	MAST1	54	PIK3CA	0	PRP4	90
BUB1	11	ERBB3	49	MEK5	6.8	PIK3CB	12	RIPK1	100
CDC2L1	87	ERK3	100	MKK7	21	PIK3CD	0	RIPK4	48
CDK11	100	ERK4	86	MTOR	0.4	PIK3CG	0	TIE1	74
CDK8	40	ERN1	85	NEK10	79	PIK4CB	0.45	TNNI3K	48
CDKL1	89	GAK	10	PCTK2	82	PIP5K1A	38	YANK1	100

[a]**15d** was screened at the concentration of 10 μM in a panel of 50 kinases, and results for primary screen binding interactions are reported as "% Ctrl", where lower numbers indicate stronger hits. % Ctrl calculation: % Ctrl = (test compound signal - positive control signal)/(negative control signal - positive control signal) × 100, negative control = DMSO (100% Ctrl), positive control = control compound (0% Ctrl).

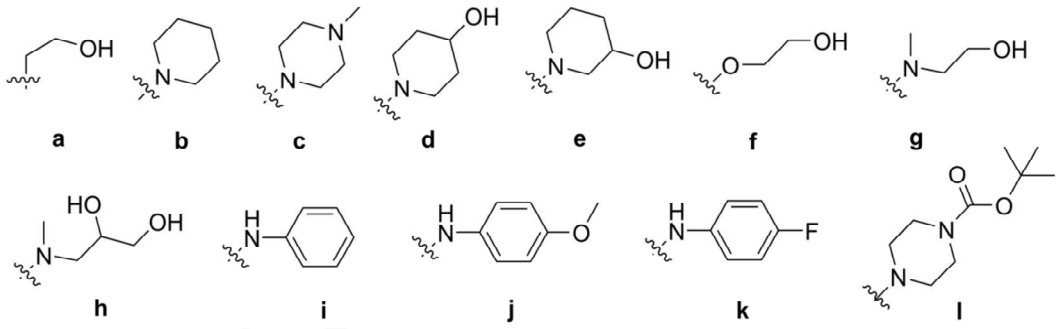
[b]Gene symbol of kinases.



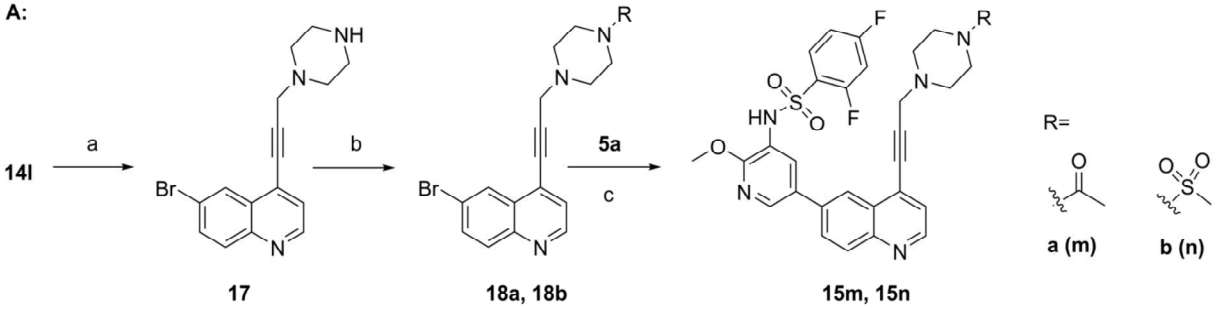
5a: R_1 = H, R_2 = F, R_3 = F; **5b**: R_1 = H, R_2 = F, R_3 = H; **5c**: R_1 = F, R_2 = H, R_3 = H;
5d: R_1 = H, R_2 = CF_3, R_3 = H; **5e**: R_1 = H, R_2 = OCF_3, R_3 = H; **5f**: R_1 = H, R_2 = CH_3, R_3 = H



13a-l, 14a-l, 15a-k R =

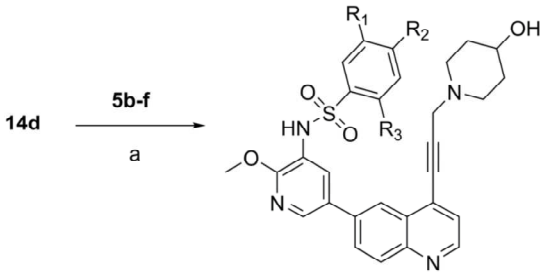


A:



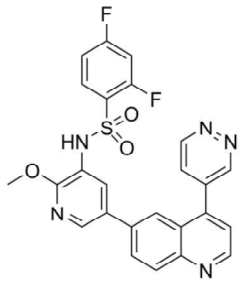
17 **18a, 18b** **15m, 15n**

B:

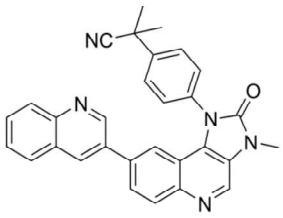


19a-e

19a : R_1 = H, R_2 = F, R_3 = H;
19b : R_1 = F, R_2 = H, R_3 = H;
19c : R_1 = H, R_2 = CF_3, R_3 = H;
19d : R_1 = H, R_2 = OCF_3, R_3 = H;
19e : R_1 = H, R_2 = CH_3, R_3 = H



GSK2126458



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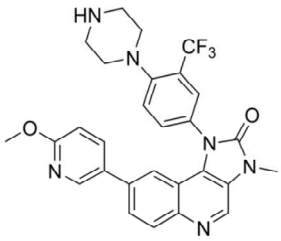

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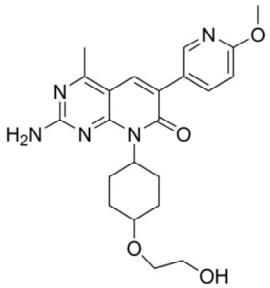

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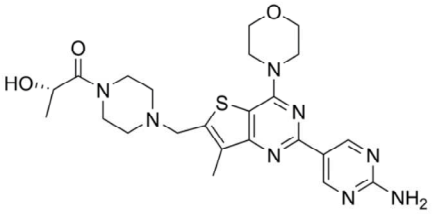
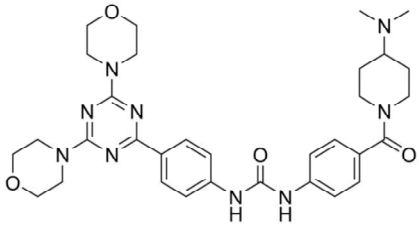
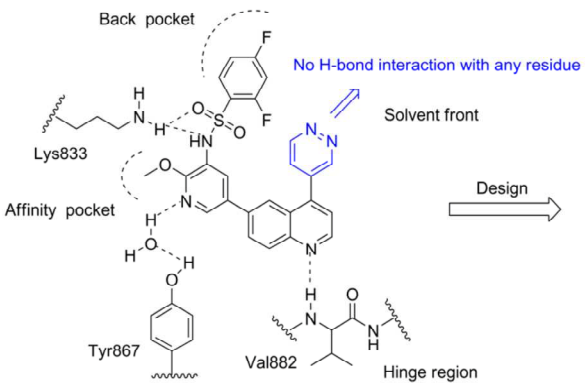


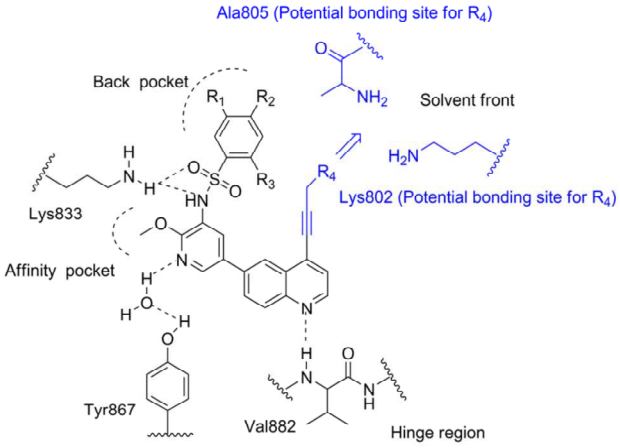
GDC-0980



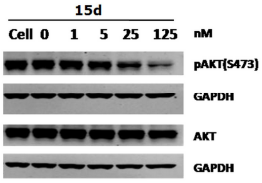
PKI-587

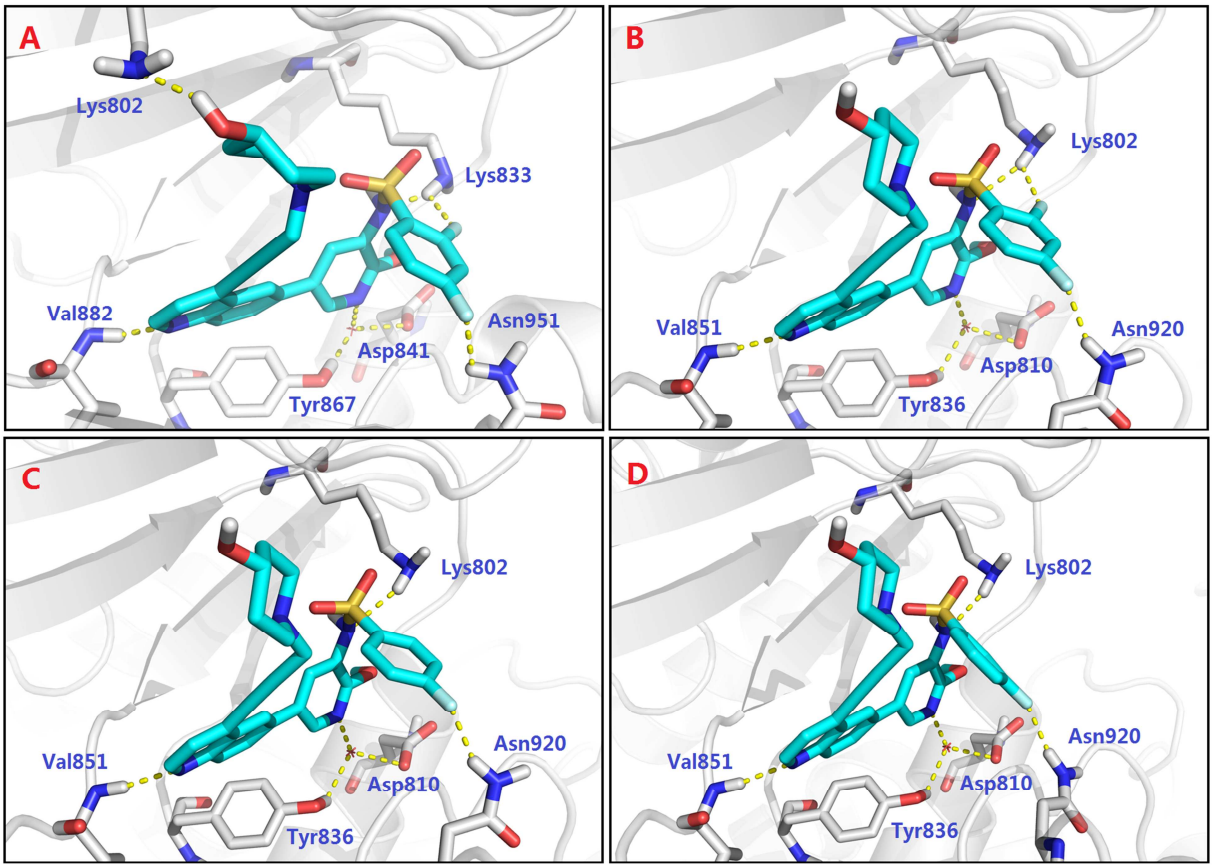


Co-crystal structure of GSK2126458 with PI3Kγ



Proposed structures and their interactions with PI3Kγ





Highlights

- Our study directly focused on new binding sites in PI3Kγ protein.
- Almost half of compounds possess high potencies less than 5 nM against PI3Kα.
- Five compounds exhibited submicromolar anti-proliferative activities.
- **15d** was the most potent compound in both PI3Kα assay and anti-proliferative assay.
- **15d** exhibited strong inhibitory effect on other class I PI3Ks, mTOR and pAkt(ser473).

Supporting Information

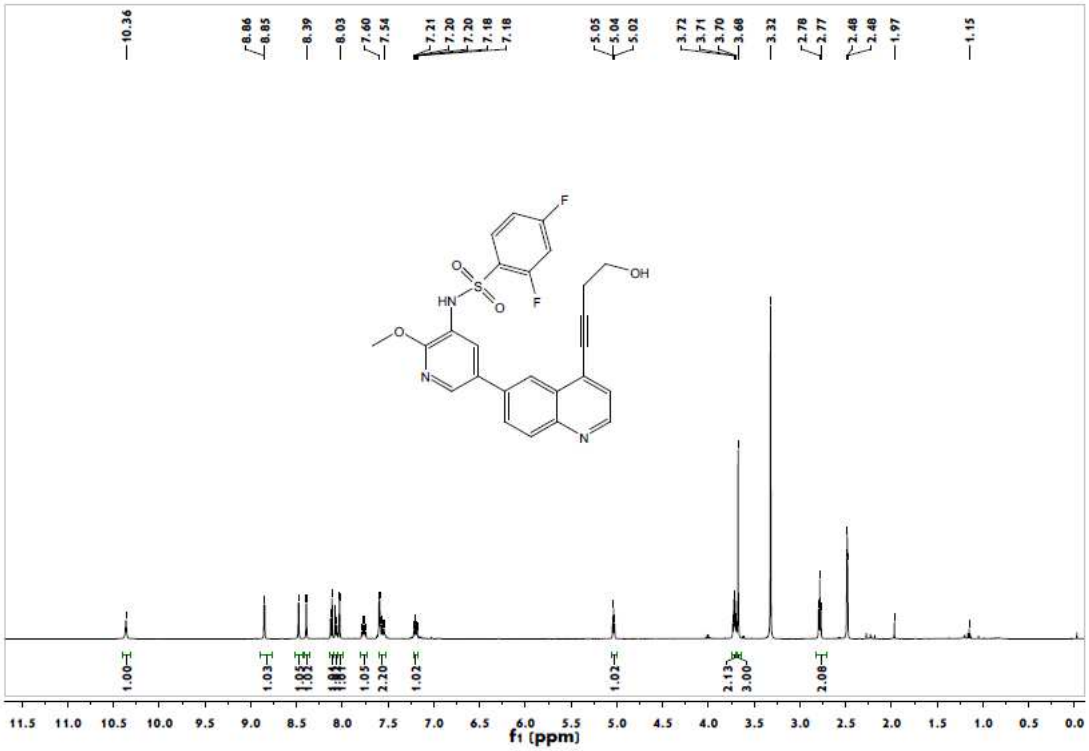
Design, synthesis and biological evaluation of novel 4-alkynyl-quinoline derivatives as PI3K/mTOR dual inhibitors

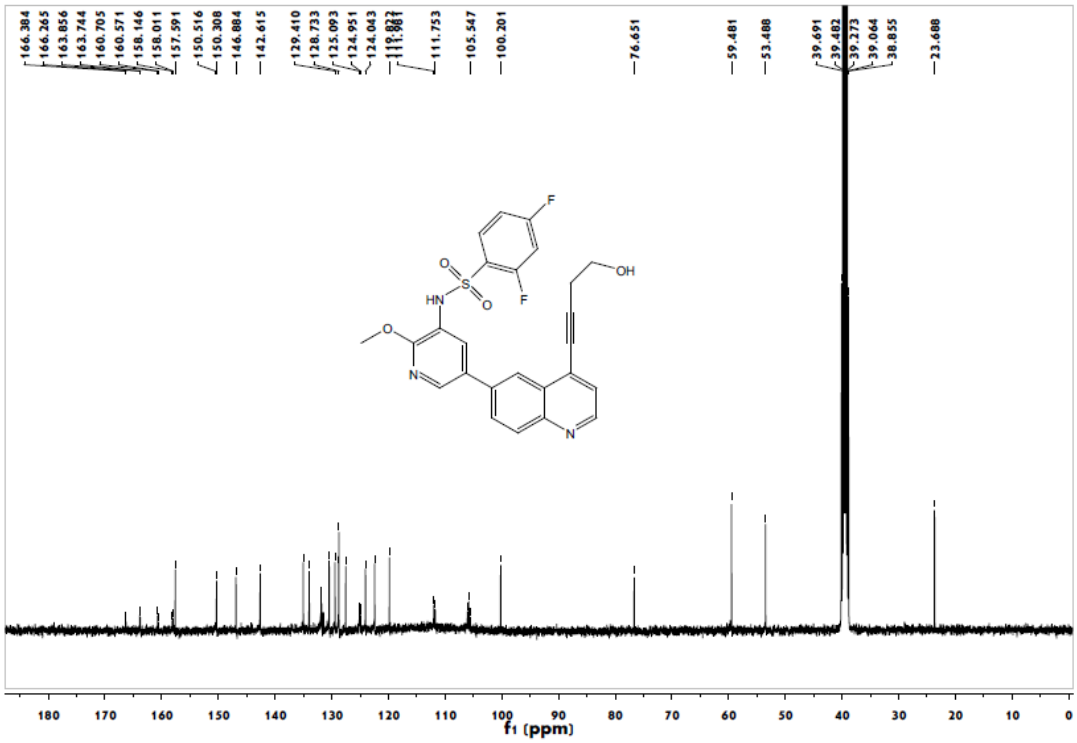
Xiaoqing Lv, Huazhou Ying, Xiaodong Ma, Ni Qiu, Peng Wu, Bo Yang, Yongzhou Hu*

Zhejiang Province Key Laboratory of Anti-Cancer Drug Research, College of Pharmaceutical Sciences, Zhejiang University, Hangzhou 310058, China

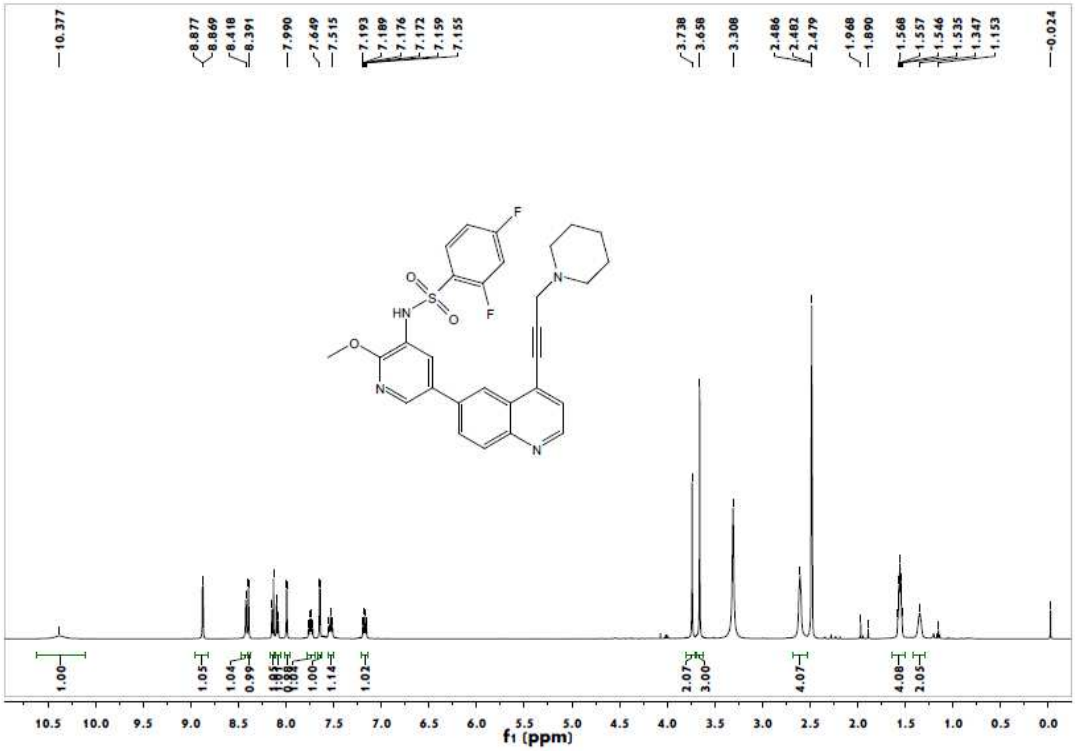
Copies of ^{1}H and ^{13}C NMR Spectra of all target compounds

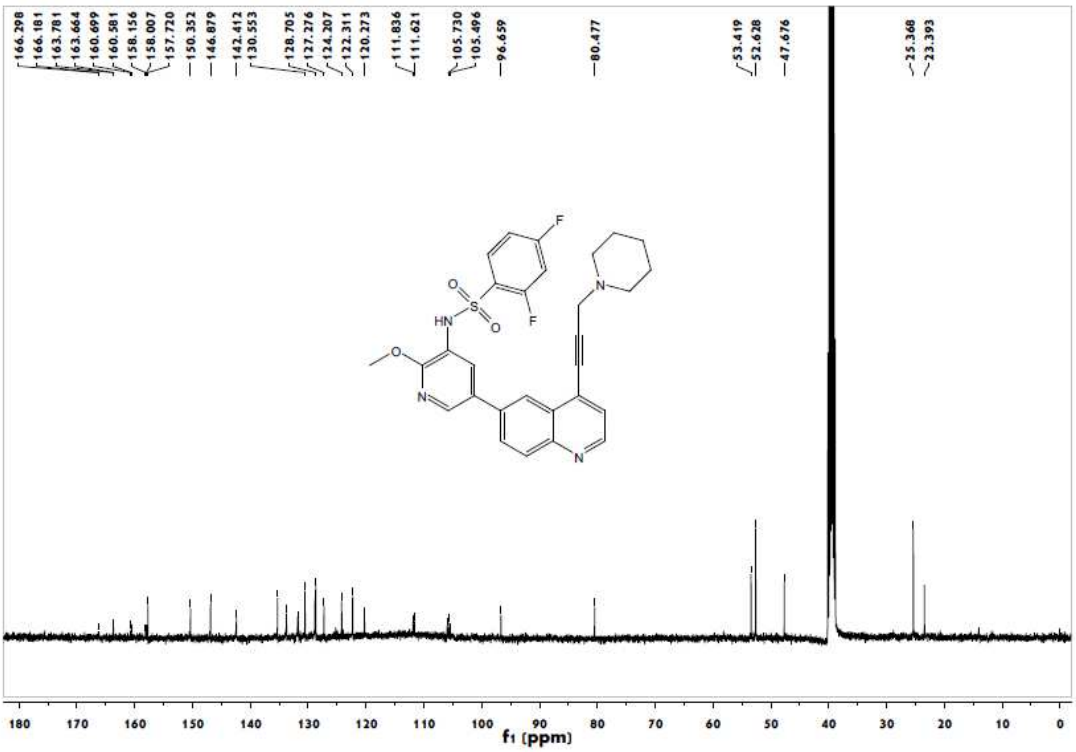
^{1}H and ^{13}C NMR of compound 15a



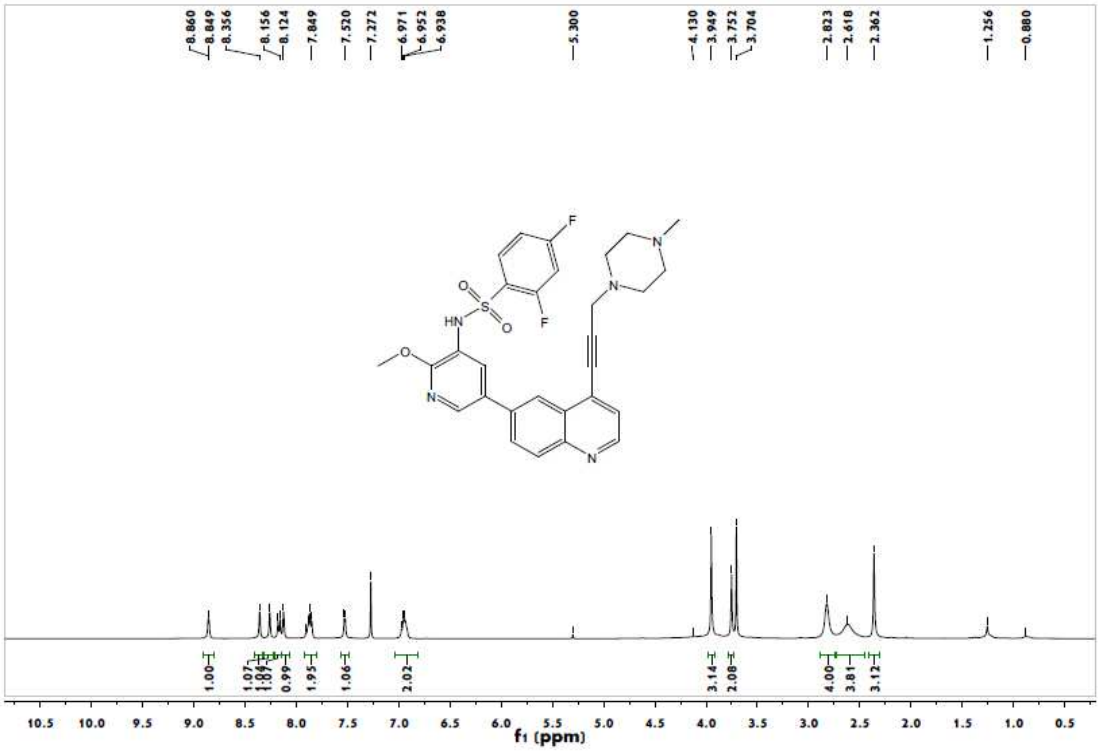


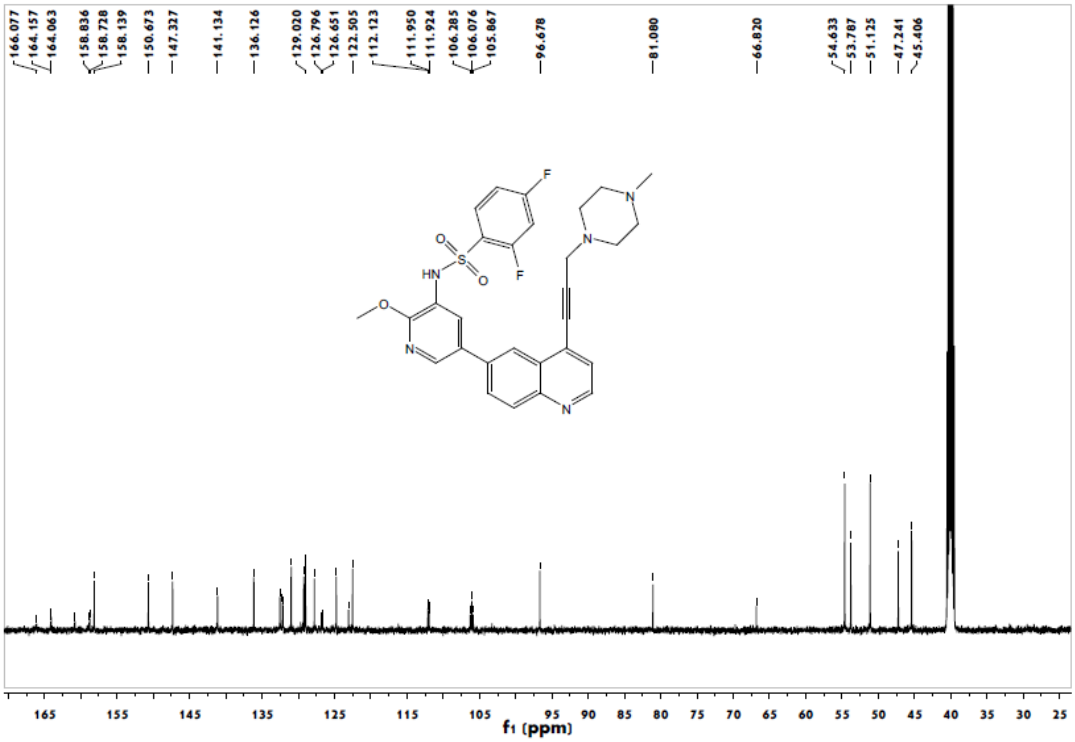
^{1}H and ^{13}C NMR of compound 15b



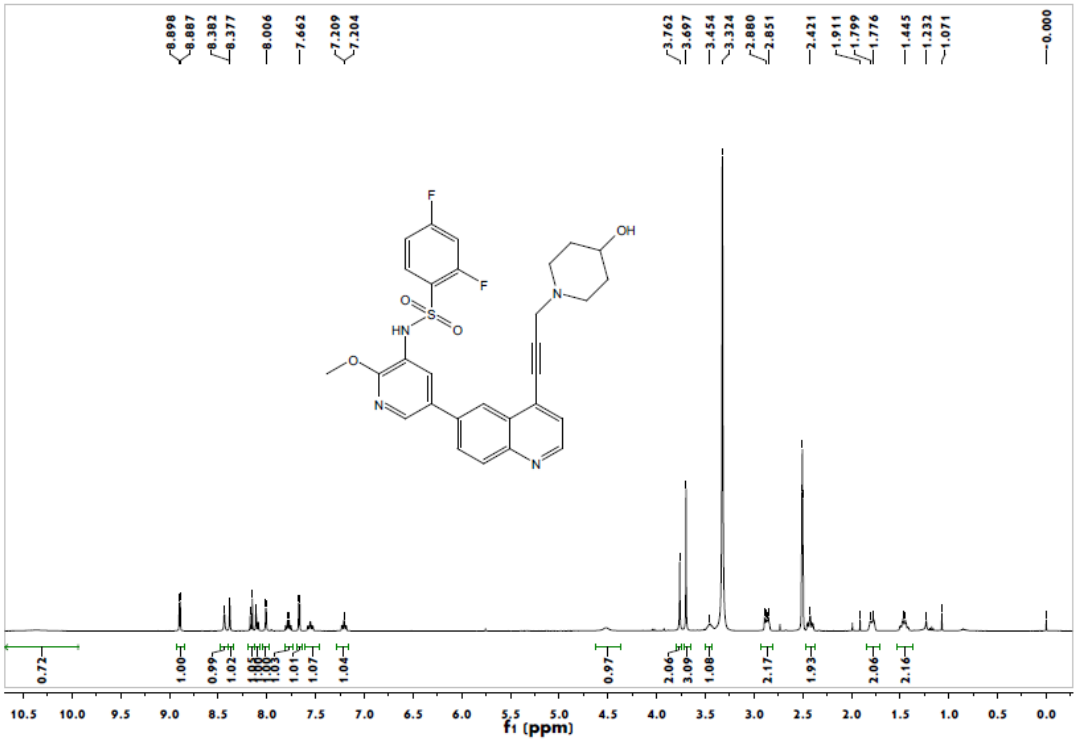


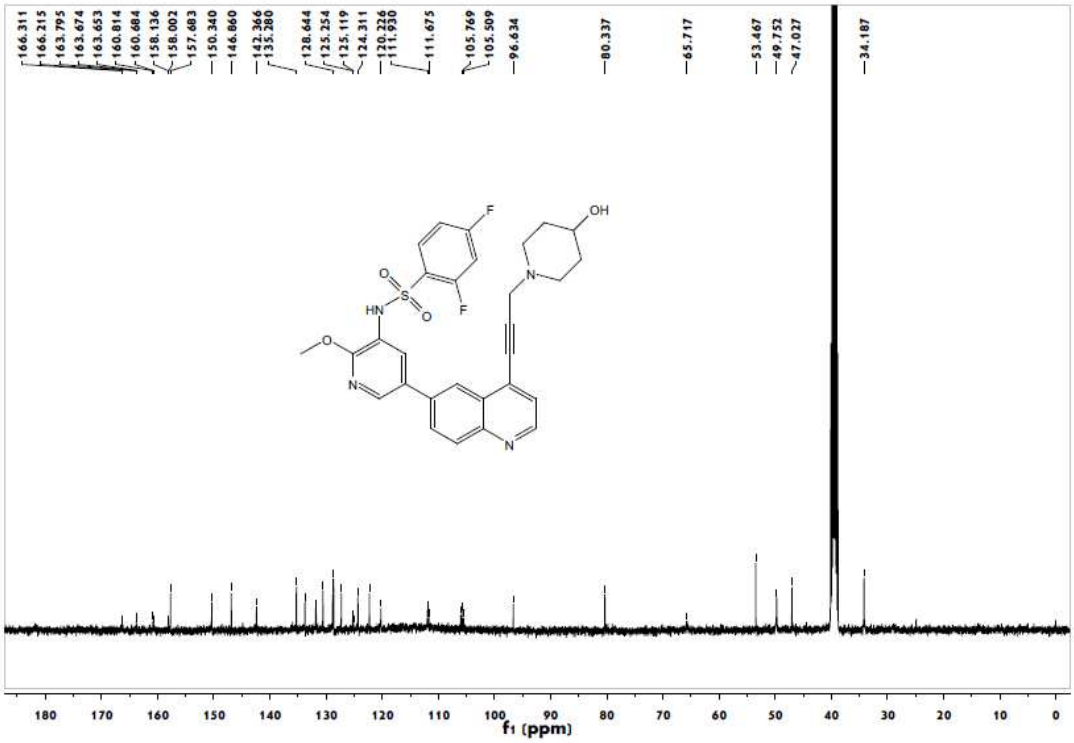
^{1}H and ^{13}C NMR of compound 15c



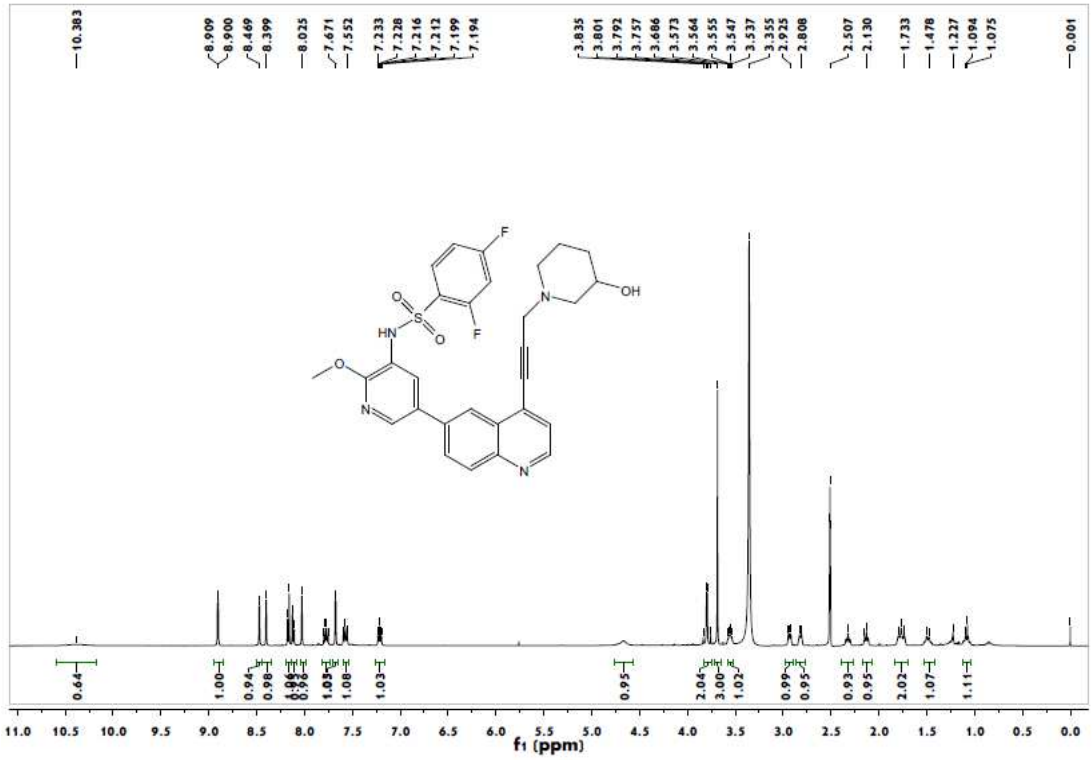


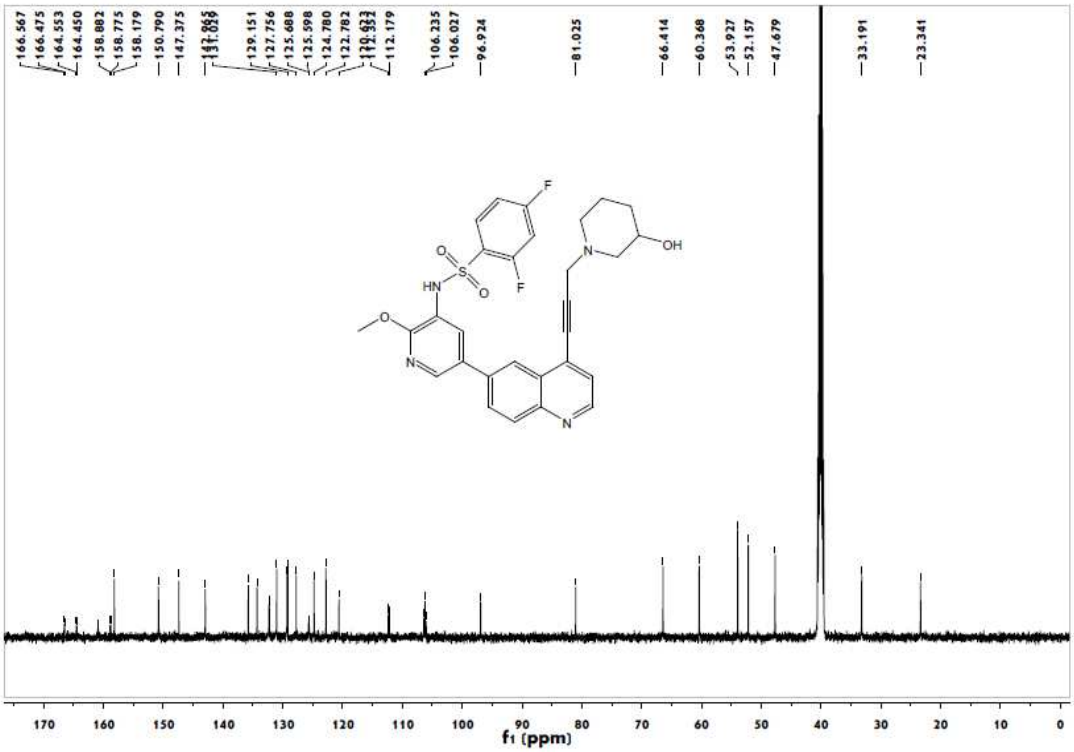
^{1}H and ^{13}C NMR of compound 15d



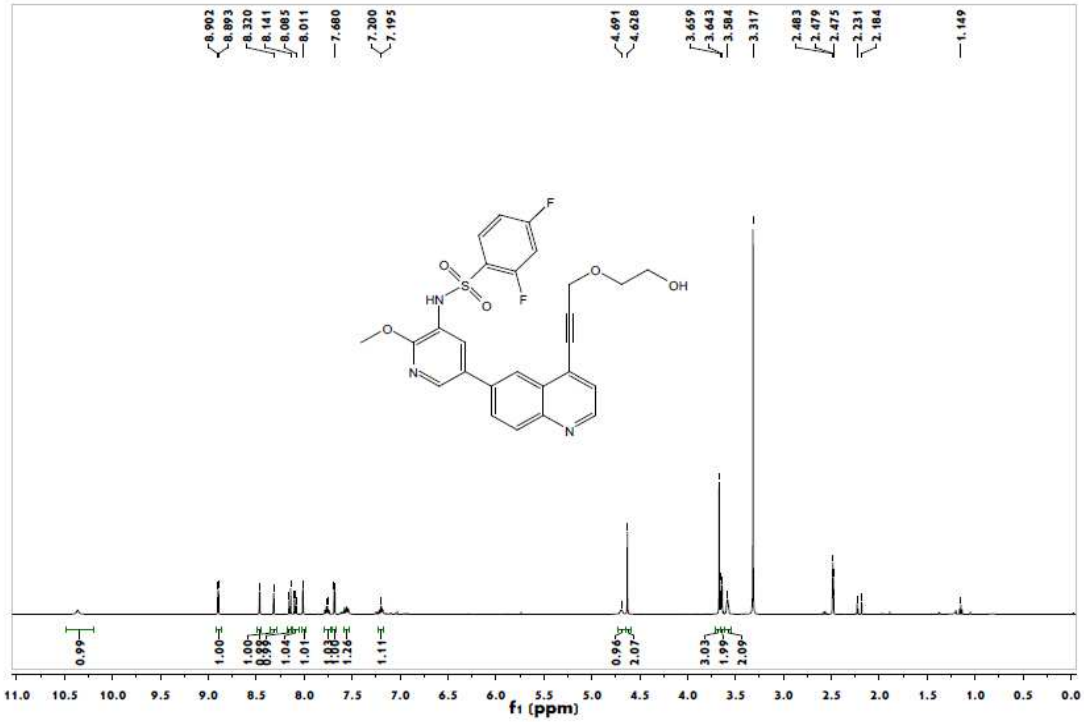


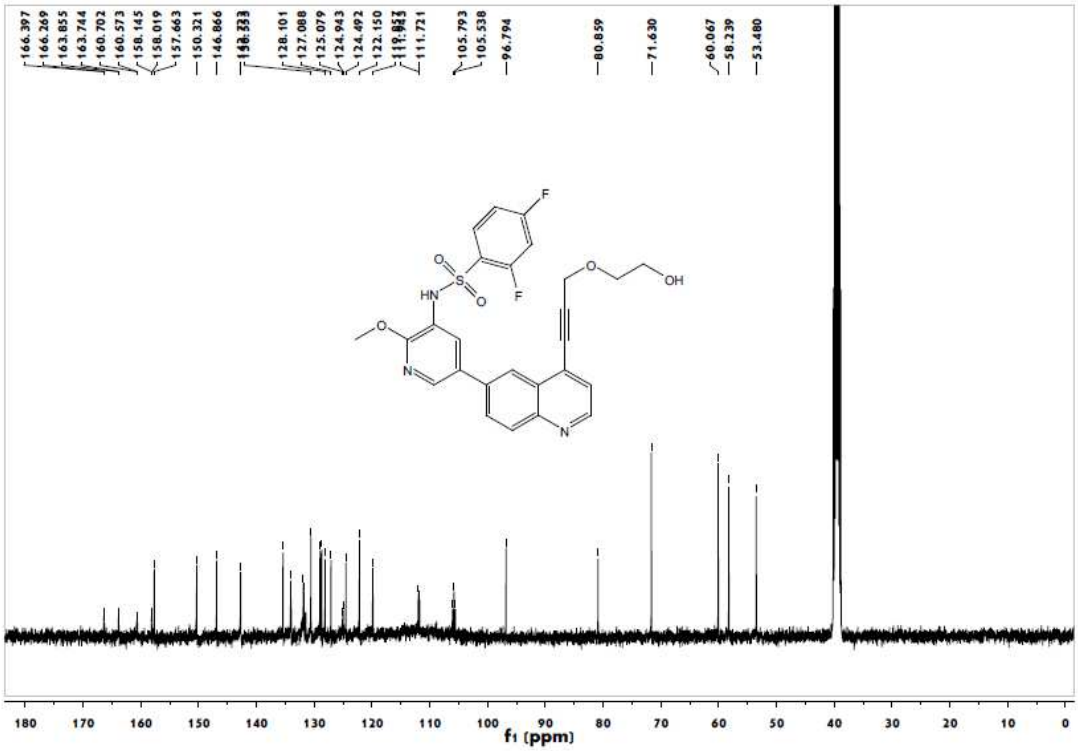
^{1}H and ^{13}C NMR of compound 15e



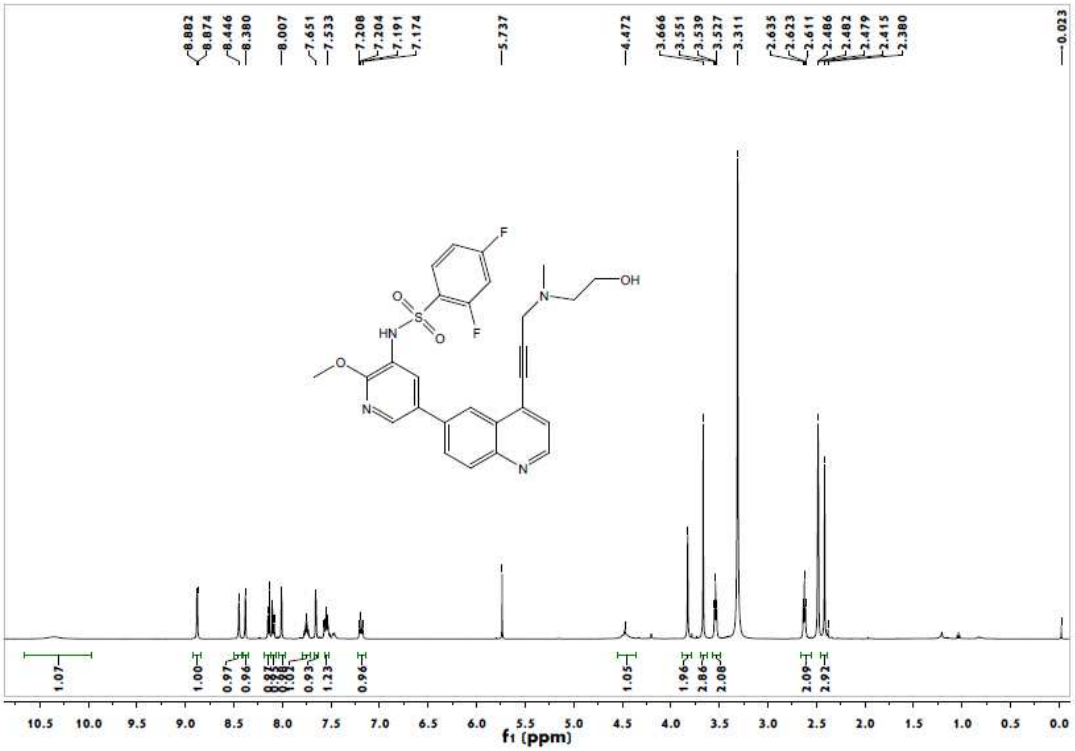


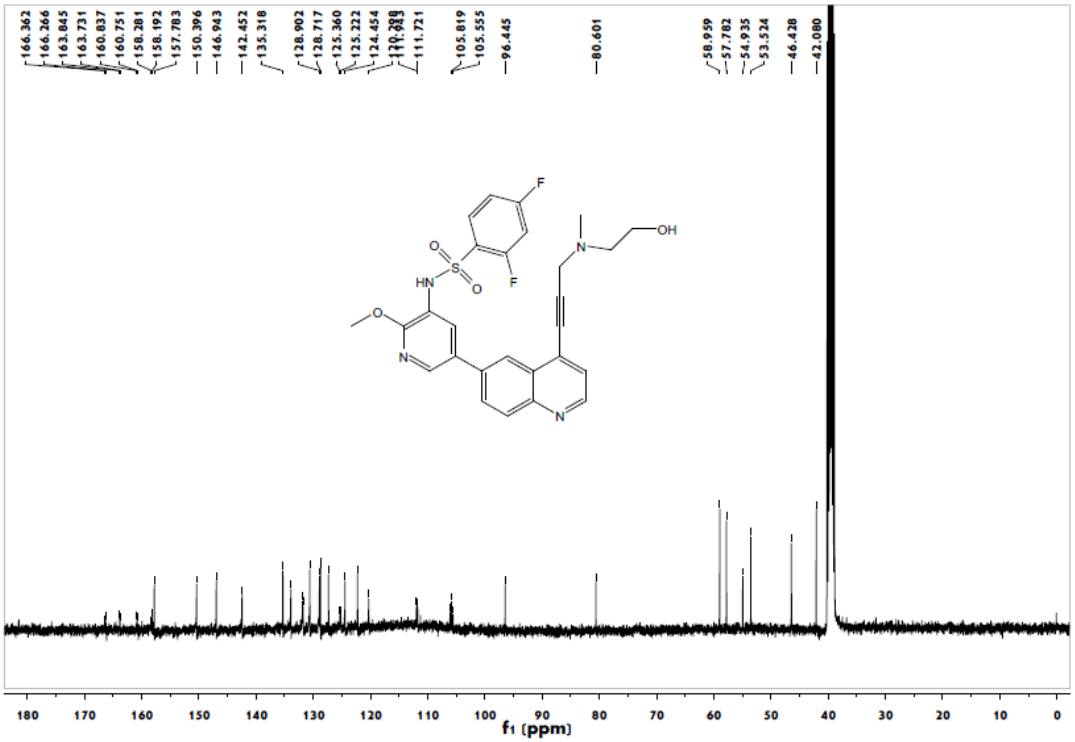
^{1}H and ^{13}C NMR of compound 15f



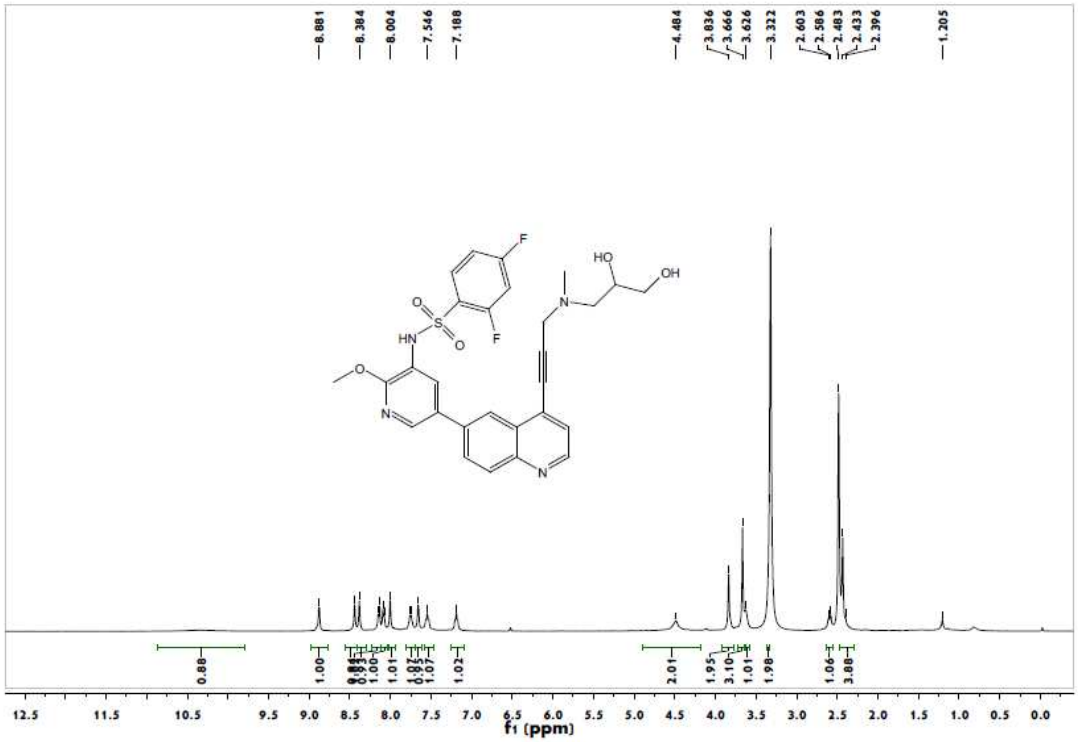


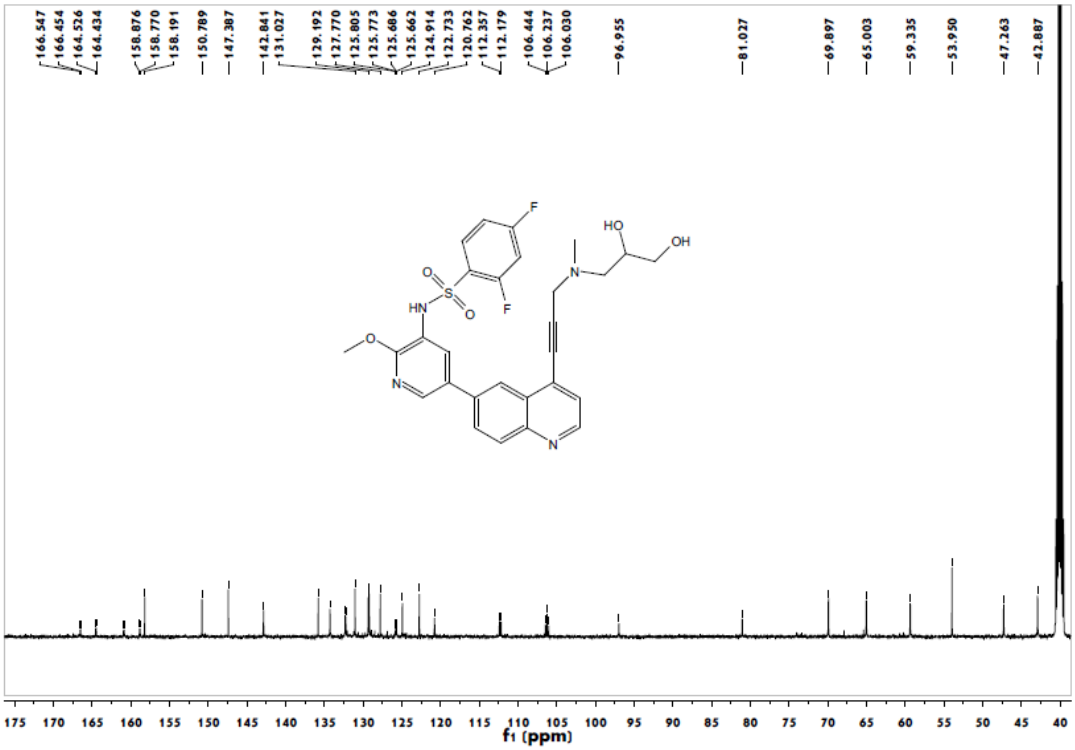
^{1}H and ^{13}C NMR of compound 15g



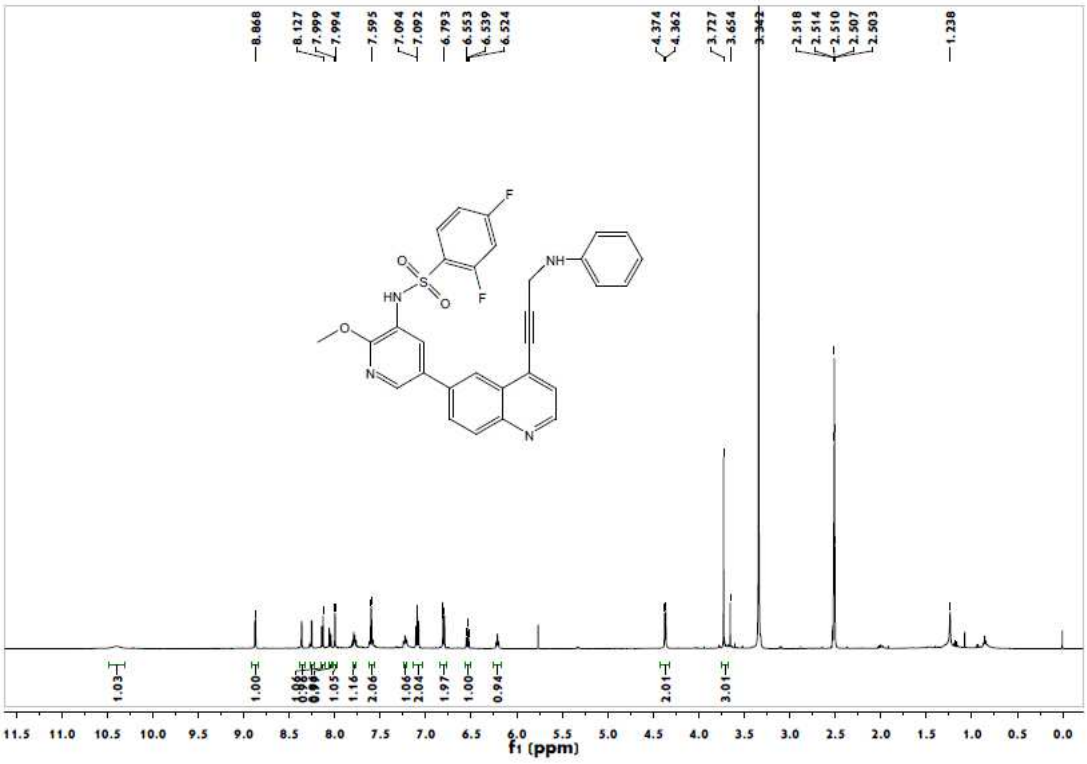


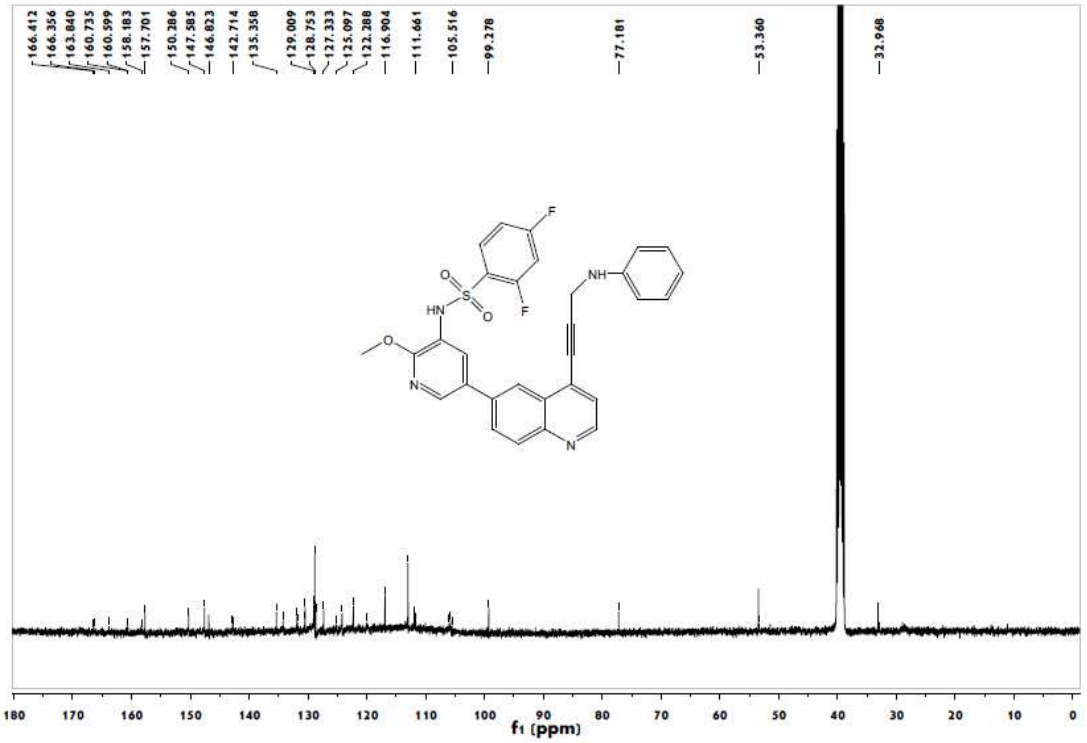
^{1}H and ^{13}C NMR of compound 15h



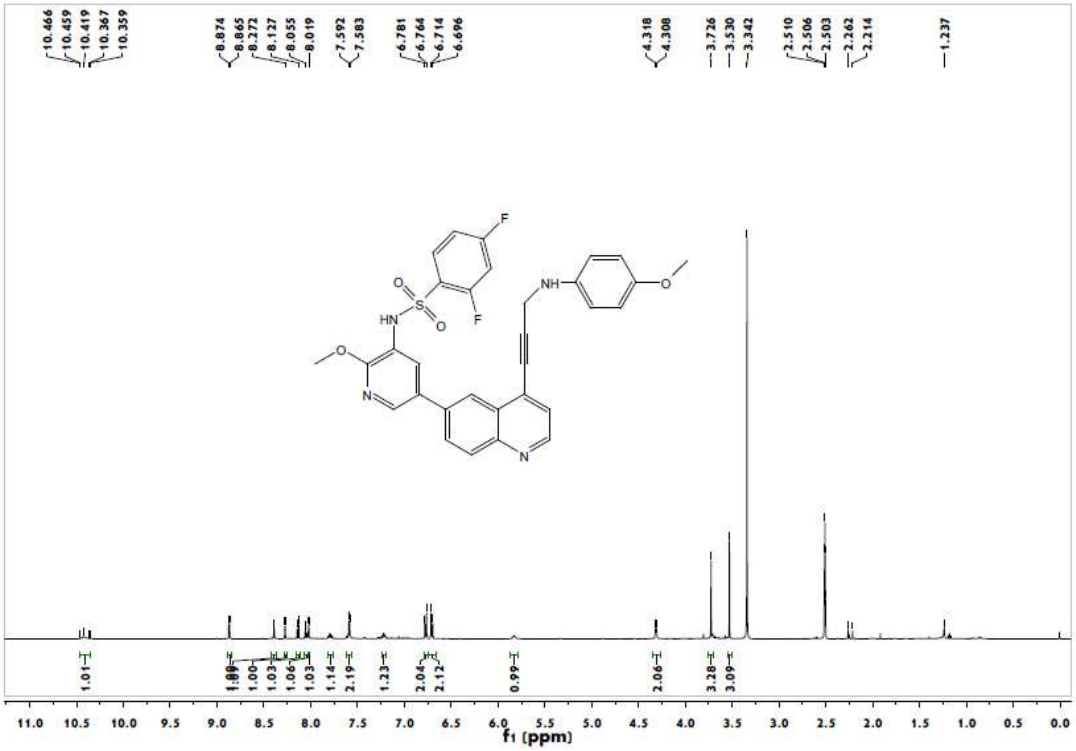


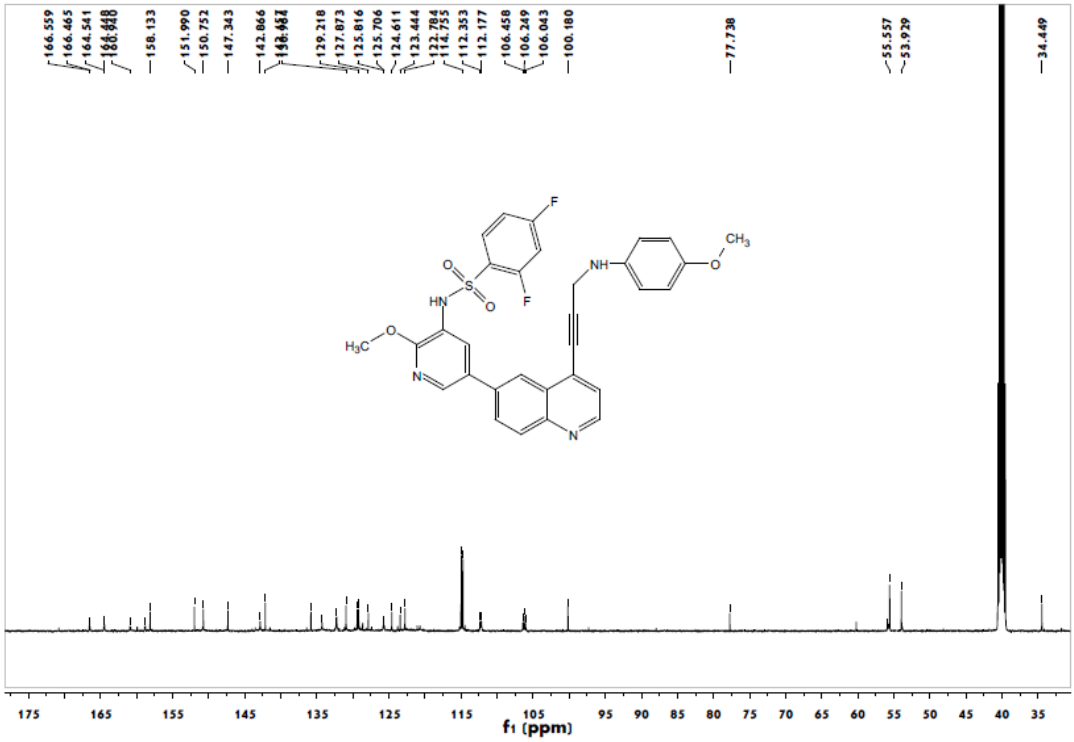
^{1}H and ^{13}C NMR of compound 15i



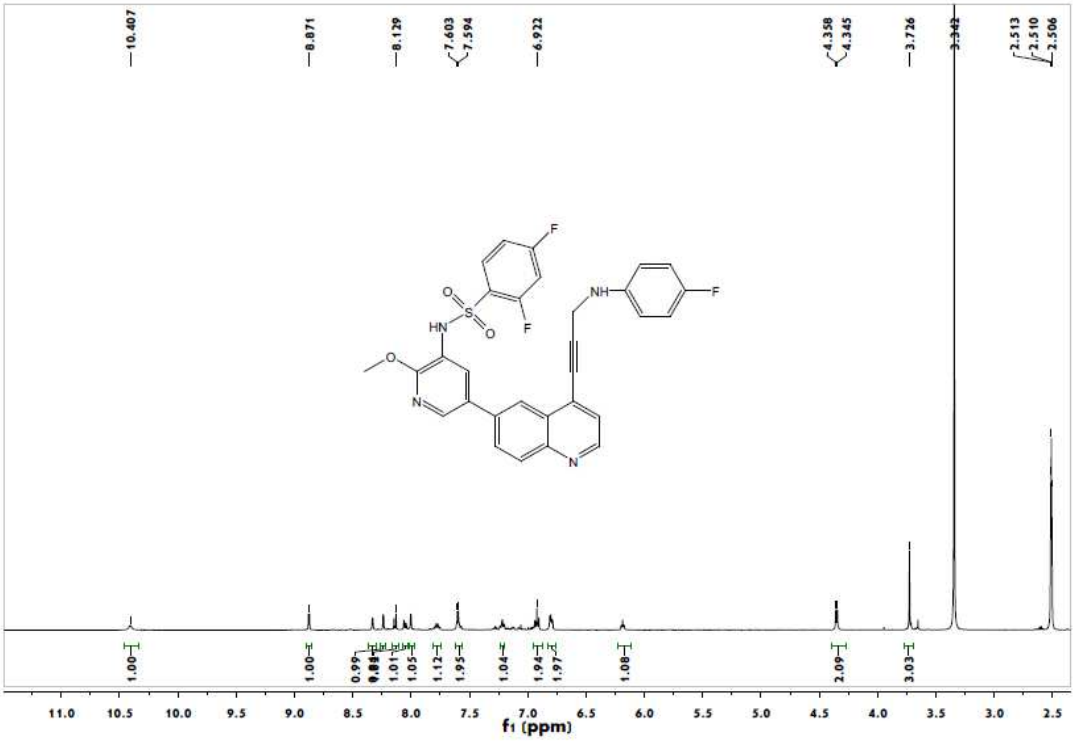


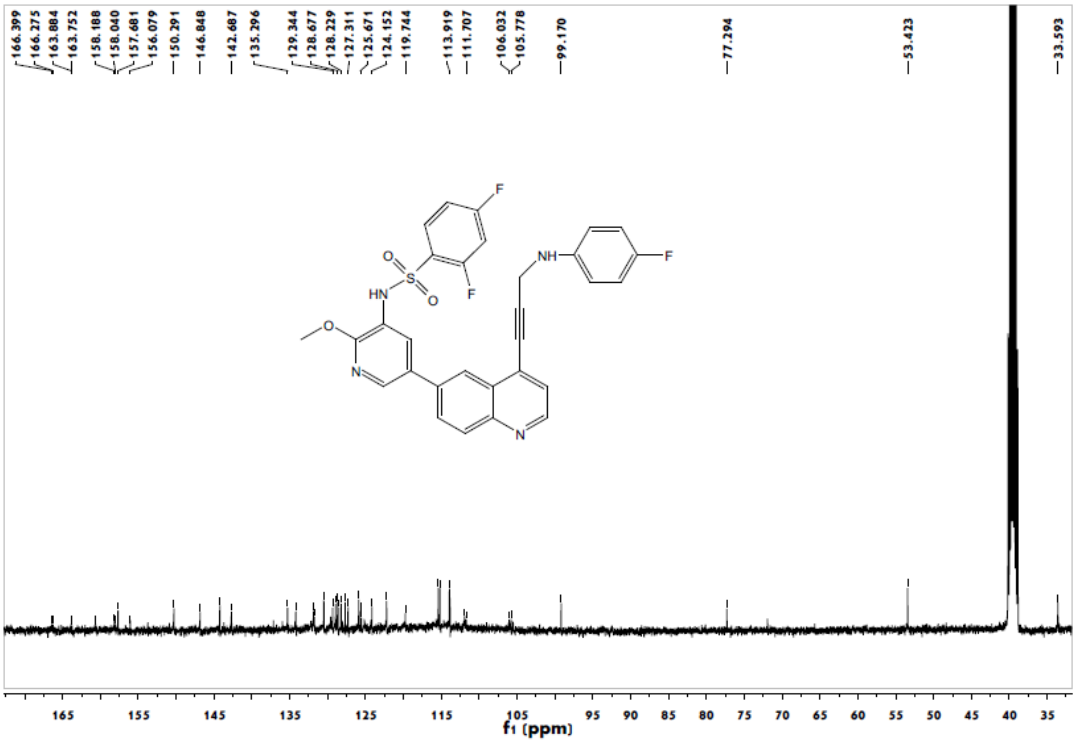
^{1}H and ^{13}C NMR of compound 15j



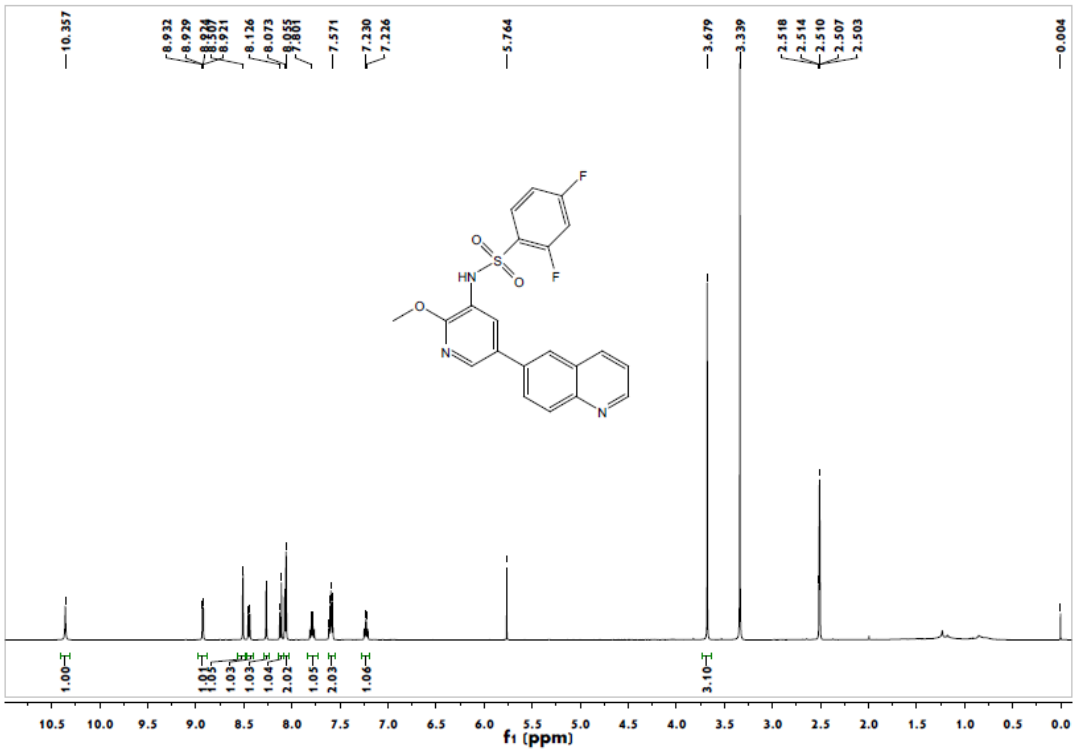


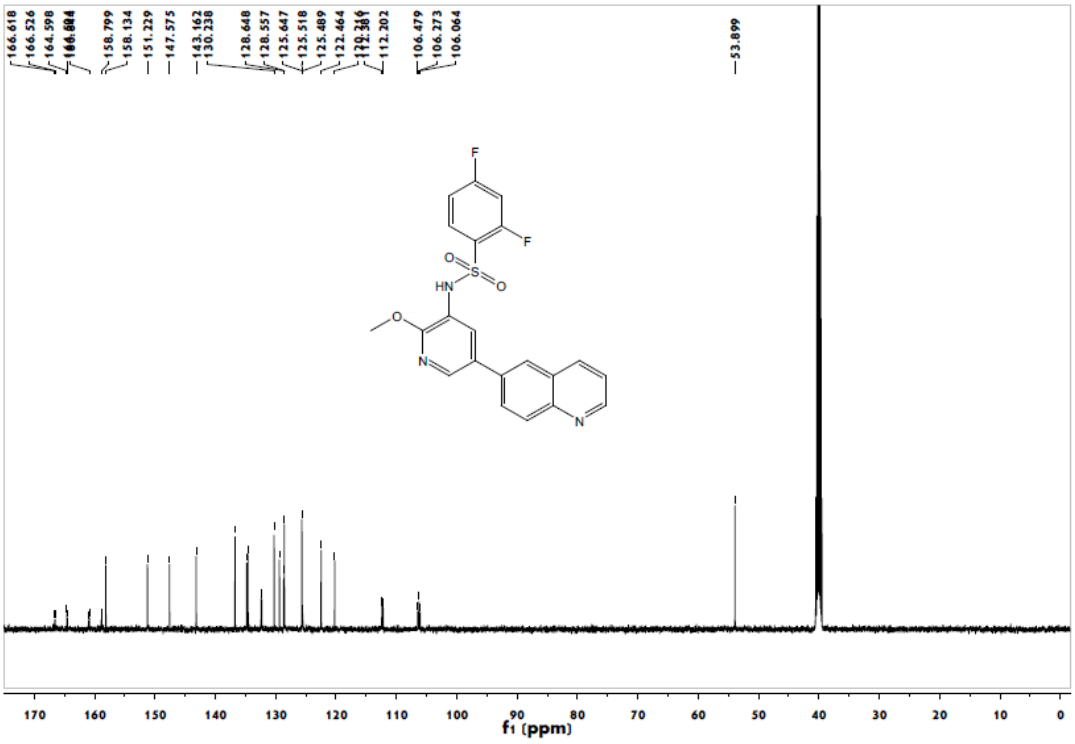
^{1}H and ^{13}C NMR of compound 15k



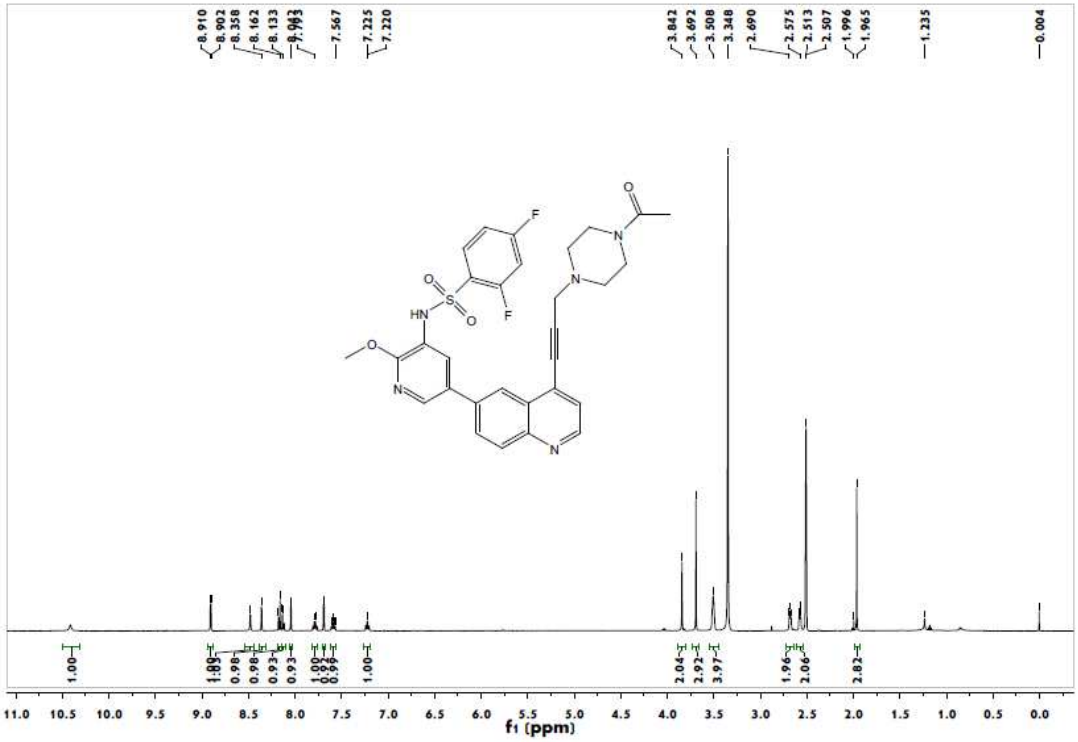


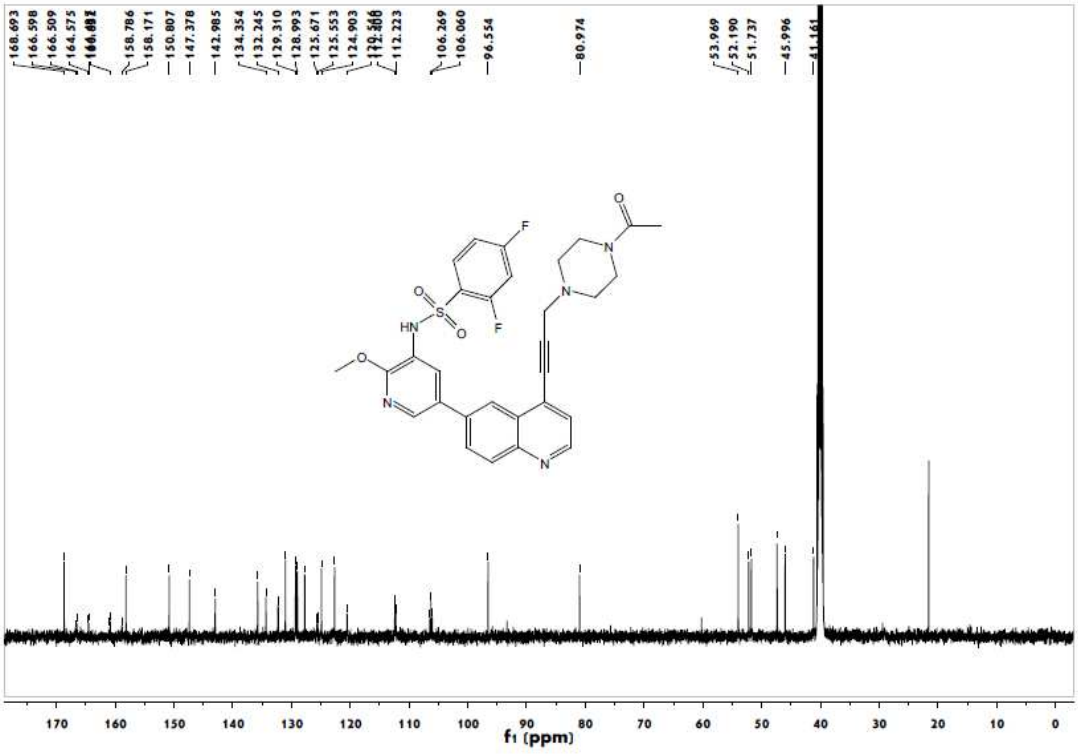
^{1}H and ^{13}C NMR of compound 15l



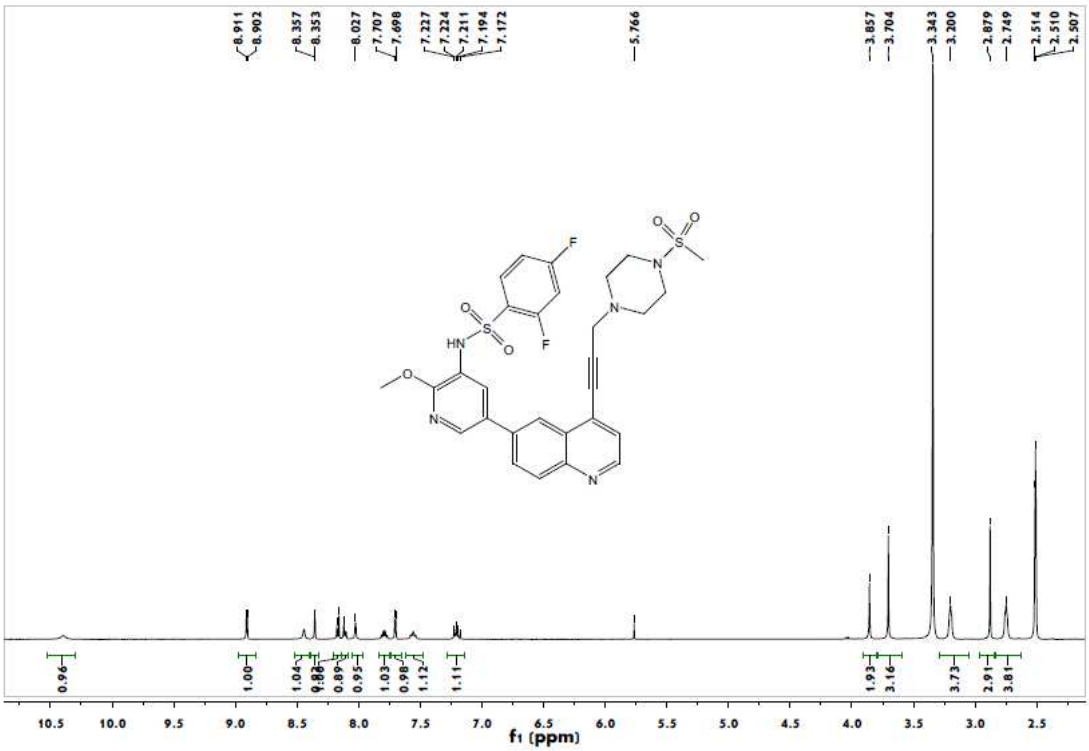


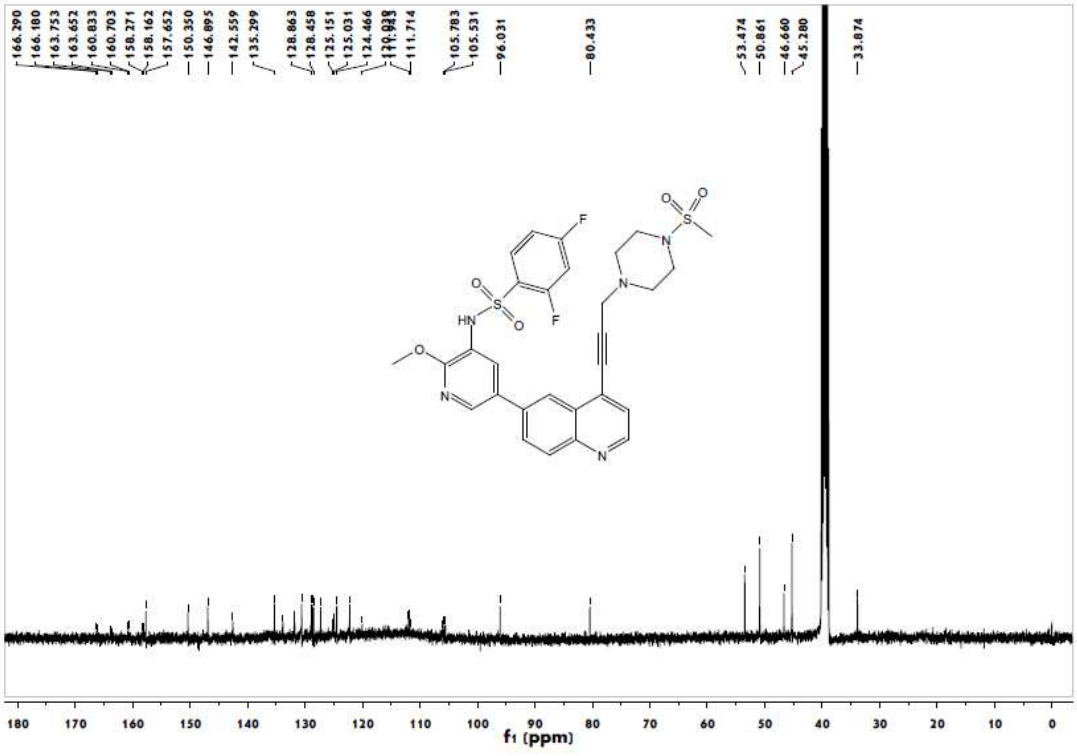
^{1}H and ^{13}C NMR of compound 15m



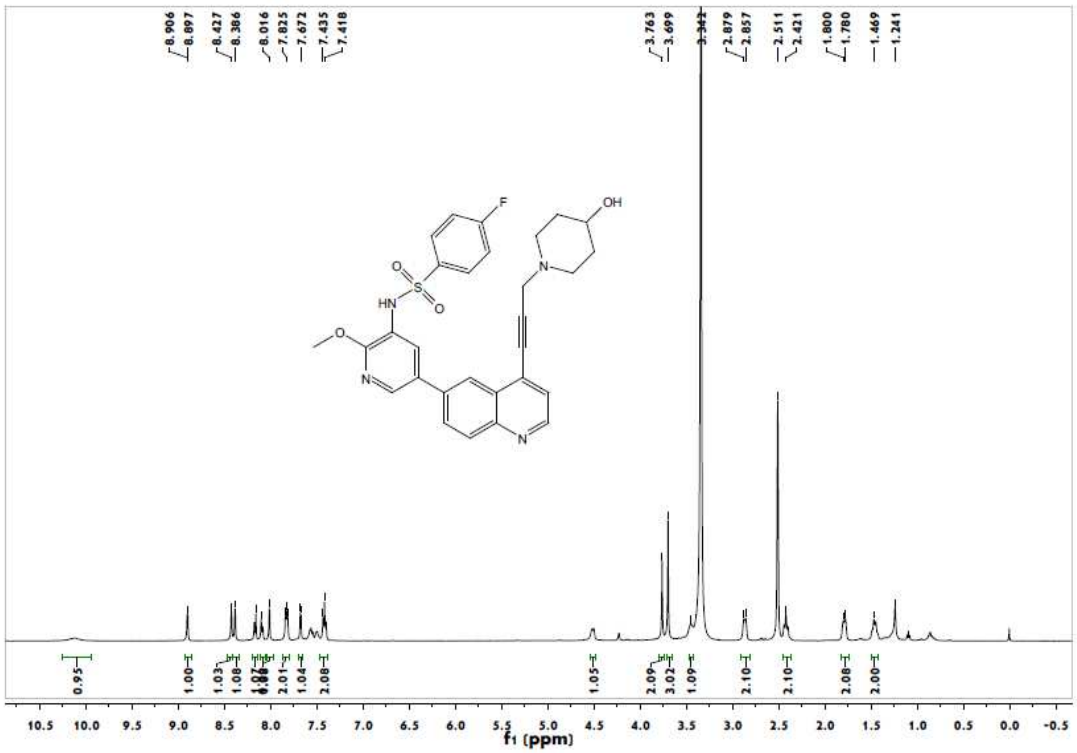


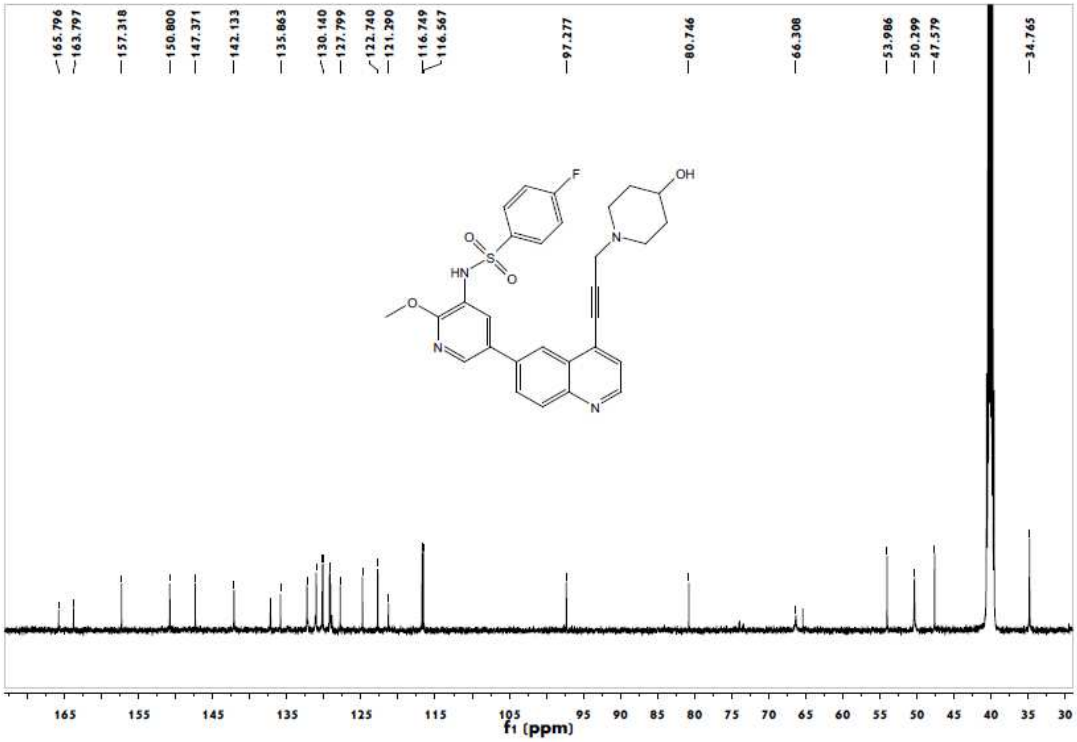
^{1}H and ^{13}C NMR of compound 15n



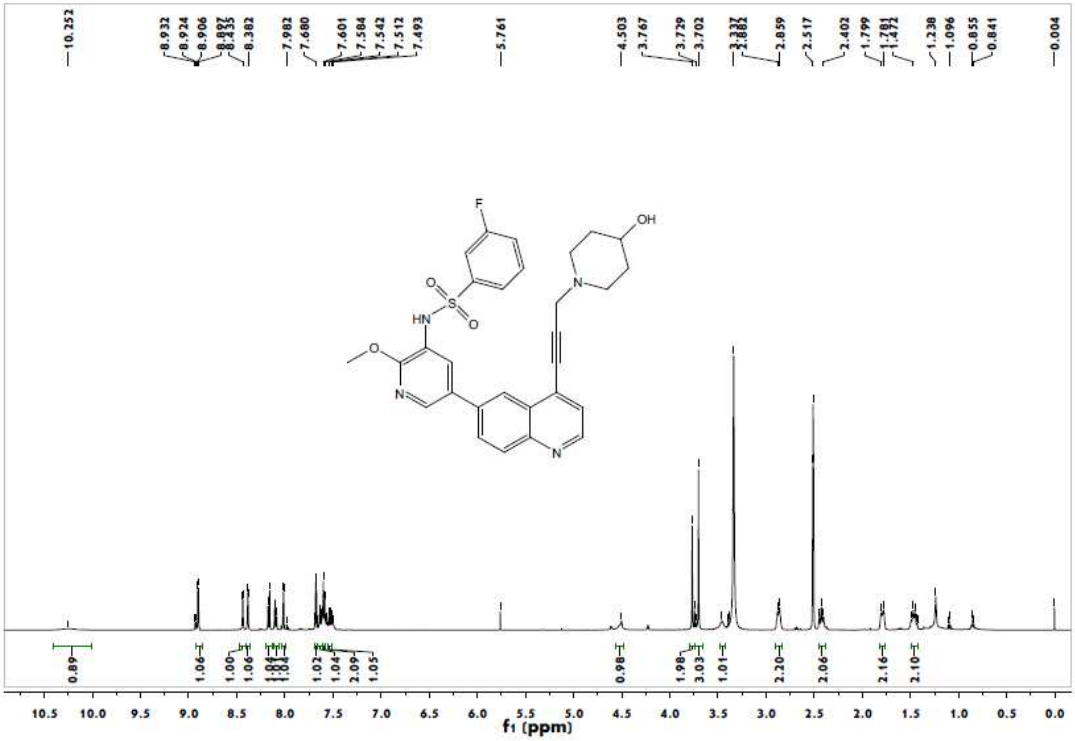


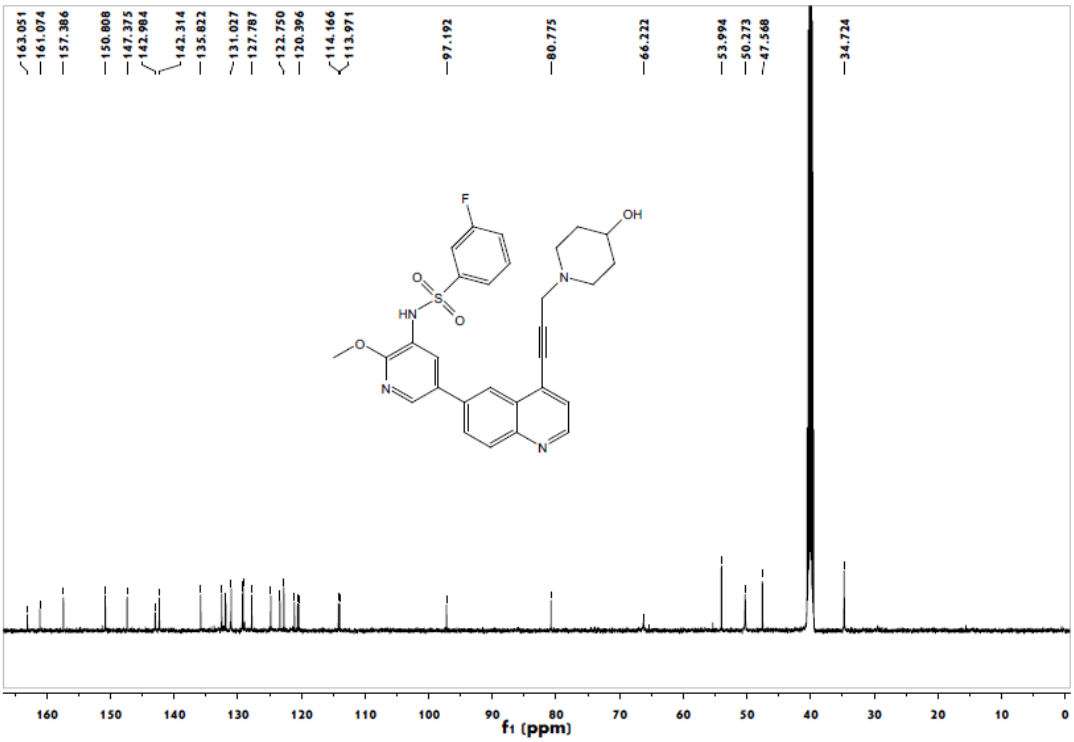
^{1}H and ^{13}C NMR of compound 19a



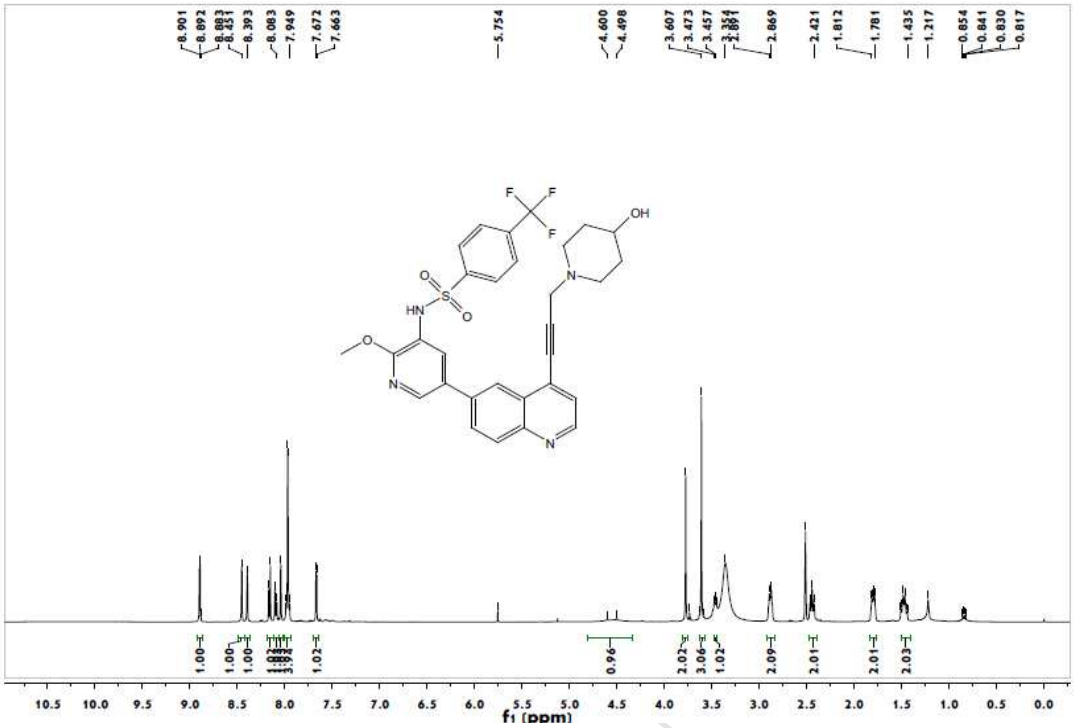


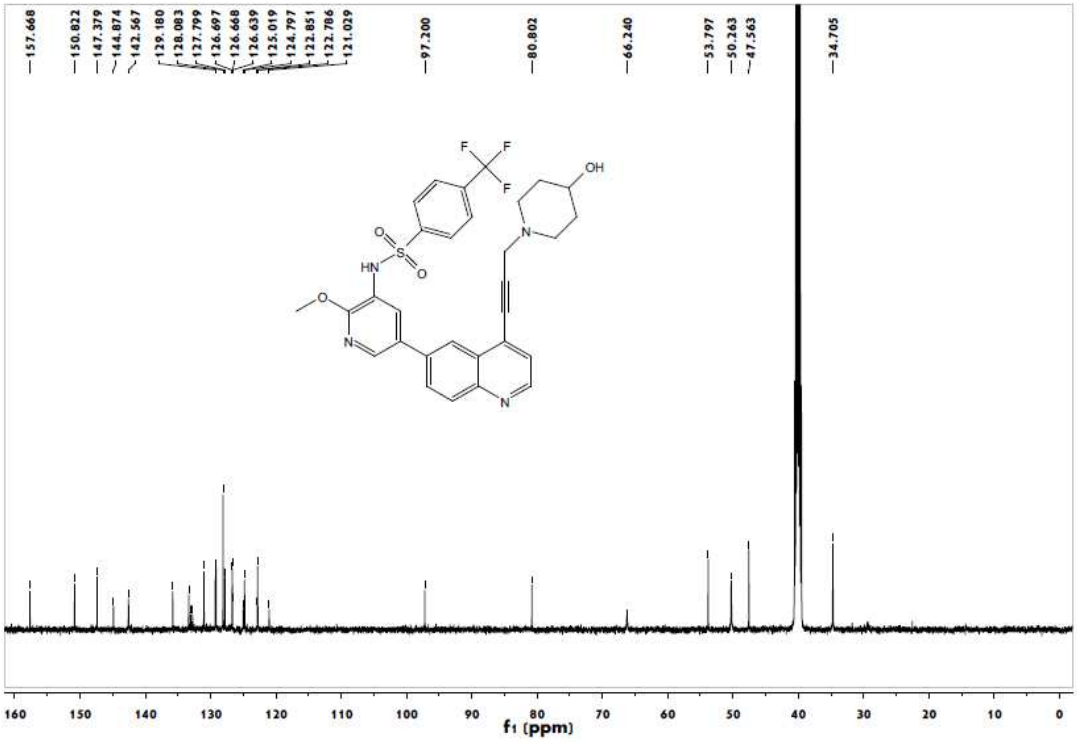
^{1}H and ^{13}C NMR of compound 19b



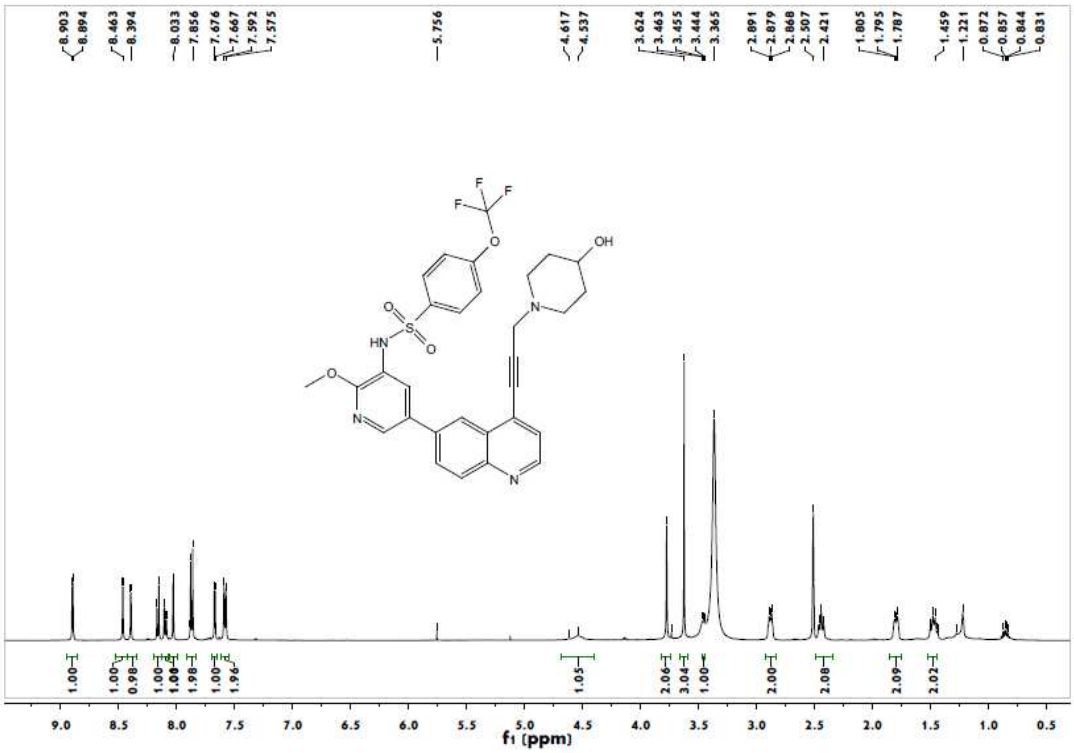


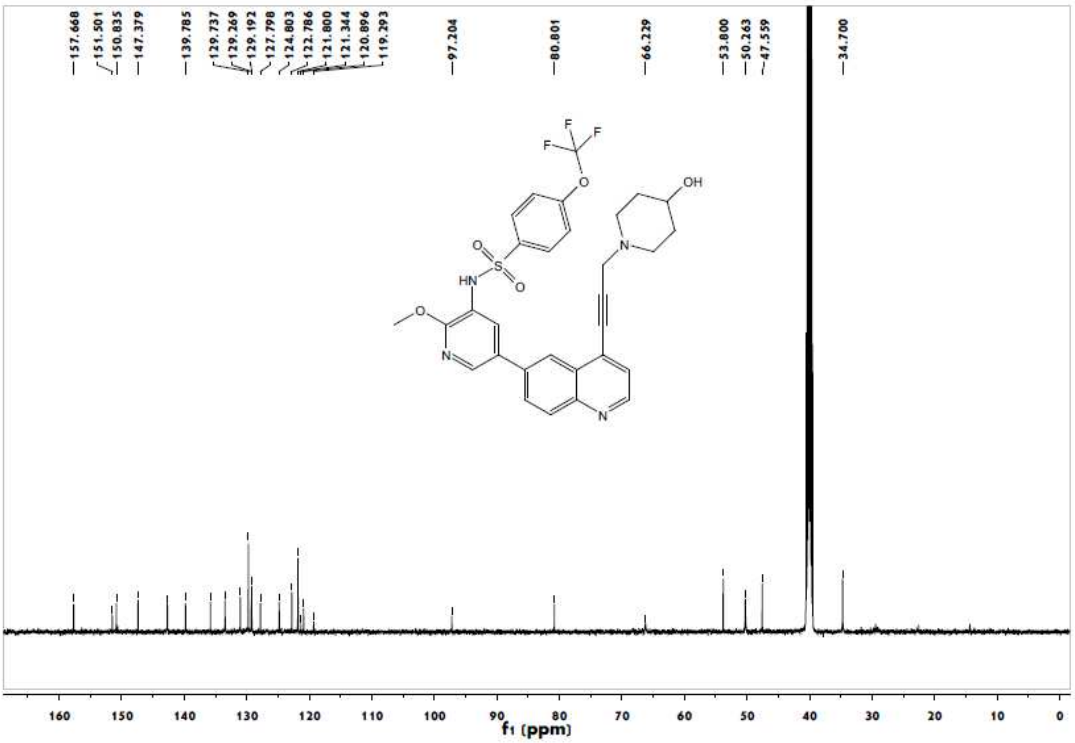
^{1}H and ^{13}C NMR of compound 19c





^{1}H and ^{13}C NMR of compound 19d





^{1}H and ^{13}C NMR of compound 19e