Discovering Some Novel 7-Chloroquinolines Carrying a Biologically Active Benzenesulfonamide Moiety as a New Class of Anticancer Agents

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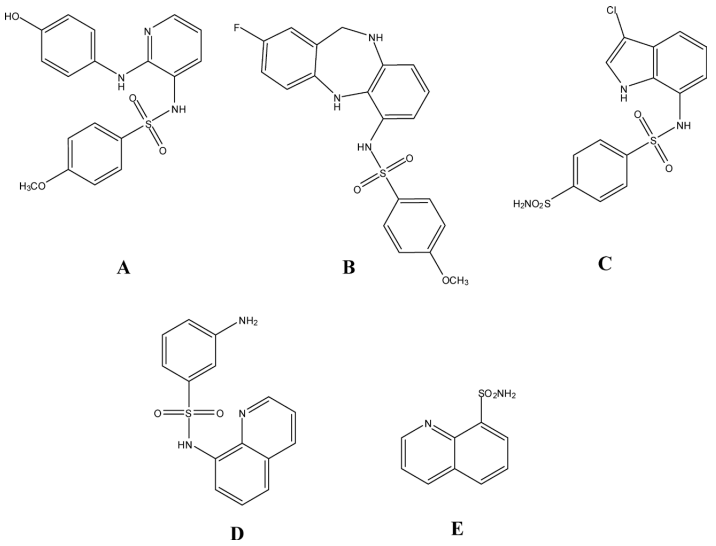
Based on the reported anticancer activity of quinolines, a new series of 7-chloroquinoline derivatives bearing the biologically active benzenesulfonamide moiety 2–17 and 19–25 were synthesized starting with 4,7-dichloroquinolne 1. Compound 17 was the most active compound with IC_{50} value 64.41, 75.05 and 30.71 μM compared with Doxorubicin as reference drug with IC_{50} values 82.53, 88.32 and 73.72 μM on breast cancer cells, skin cancer cells and neuroblastoma, respectively. All the synthesized compounds were evaluated for their *in vitro* anticancer activity on breast cancer cells, skin cancer cells and neuroblastoma cells. Most of the synthesized compounds showed moderate activity. In order to suggest the mechanism of action for their cytotoxic activity, molecular docking for all synthesized compounds was done on the active site of phosphoinositide kinase (PI3K) and good results were obtained.

Key words qunioline; sulfonamide; anticancer activity

Many quinoline derivatives possess a wide variety in pharmacological activity.[1–3] The quinoline pattern is found in a large number of natural products and drug-like compounds known as antitumor agents, such as Camptothecin,[4] Luotonin,[5] Ascididemin,[6] TAS-103,[7] Cryptolepin,[8] Indolo[2,3-*b*]quinolines.[9] In addition, sulfonamides constitute an important class of drugs with several types of pharmacological activities including antibacterial,[10] anti-carbonic anhydrase,[11] diuretic,[12] hypoglycemic,[13] antitumor[2] and antithyroid activity.[14] Some structurally novel sulfonamide derivatives have recently been reported to show substantial antitumor activity *in vitro* and/or *in vivo*. Compounds such as E7010 **A**, ER-34410 **B**, E7070 Indisulam **C**, QBS **D** and the quinoline derivative **E** are examples for antitumor sulfonamides in advanced clinical trials.[15] It has been known that aryl/heteroaryl sulfonamides may act as antitumor agents through a variety of mechanisms such as cell cycle perturbation in the G1 phase, disruption of microtubule assembly, angiogenesis inhibition, and functional suppression of the transcriptional activator NF-Y. Moreover, following an extensive evaluation, numerous sulfonamides were found to act as carbonic anhydrase (CA) inhibitors.[16–18] Recently, combination of quinoline nuclues with sulfonamide moieties has received a great attention in seeking for novel anticancer agents.[19] Several quinoline sulfonamide derivatives showed potent anticancer activity as phosphoinisitol kinase (PI3K) inhibitors.[19] In view of the above mentioned findings and as a part of our research effort to explore novel anticancer heterocyclic compounds[20,21] we have synthesized a new series of chloroquinoline derivatives having biologically active sulfonamide moiety as analogues for compounds **A**–**E**. The cytotoxic activity of the newly synthesized compounds were evaluated through *in vitro* antitumor screening on several cell



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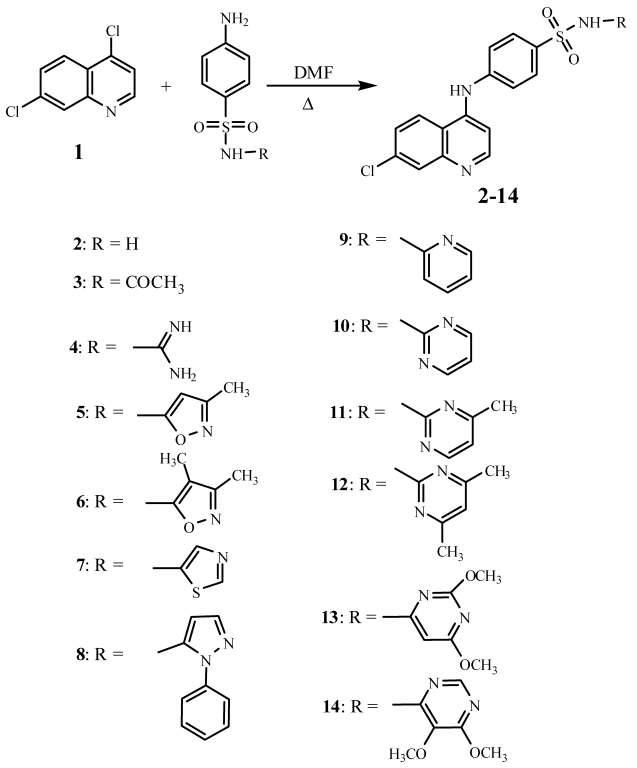

Chart 1. Synthetic Pathways for Compounds **2**–**14**

lines. Moreover, as a trial to suggest the mechanism of action of these compounds virtual docking on the active site of PI3K was performed.

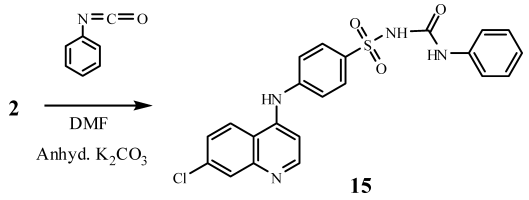

Chart 2. Synthetic Pathway for Compound **15**

Results and Discussion

Chemistry Several compounds were designed with the aim of exploring their anticancer properties (Charts 1–3). The present work reports the possible utility of 4,7-dichloroquinoline **1** in the synthesis of 7-chloroquinolinesulfonamide derivatives **2**–**17** and **19**–**25**. Quninolinesulfonamide derivatives **2**–**14** were obtained in good yield by the reaction of compound **1** with several sulfonamides in dry *N*,*N*-dimethylformamide (DMF), whereas, conducting this reaction in the presence of anhydrous potassium carbonate[22] afforded the corresponding sulfonamide derivative **16**. The structures of **2**–**14** and **16** were supported by elemental analysis and spectral data. The IR spectra of compounds **2**–**14** revealed the presence of bands for NH, CH aromatic, C=N, SO_2 and C–Cl. IR spectrum for compound **16** showed bands at 3454, 3389 and 3244 cm^{-1} (NH, NH_2), 1627 cm^{-1} (C=N), 1363, 1136 cm^{-1} (SO_2), 808 cm^{-1} (C–Cl). ^{1}H-NMR spectra of compounds **2**–**14** in (DMSO-d_6) indicated the presence of a doublet at 6.5–8.6 ppm which could be assigned to 2CH of pyridine and a singlet at 10.2–10.7 ppm for NHph. ^{1}H-NMR spectrum of compound **16** exhibited singlet at 5.7 ppm due to NH_2 group. On the other hand, 4-(7-chloroquinolin-4-ylamino)-*N*-(phenylcarbamoyl)-benzenesulfonamide **15** was obtained in a good yield *via* reaction of compound **2** with phenyl isocyanate in dry DMF in the presence of anhydrous potassium carbonate. Compound **15** was confirmed by elemental analysis as well as spectral data. The IR spectrum for compound **15** revealed the presence of bands corresponding to (NH), (C=O), (C=N), (SO_2) and (C–Cl) groups. Its ^{1}H-NMR spectrum in (DMSO-d_6) showed singlet at 8.4 ppm for NHCO group. When compound **16** was reacted with phenyl isocyanate in dry DMF the corresponding ureido derivative **17** was obtained. The IR spectrum of compound **17** revealed the presence of bands corresponding to (NH), (C=O), (SO_2) groups. Its ^{1}H-NMR spectrum in (DMSO-d_6) showed signals at 9.1, 9.2 ppm assigned to 2NH of ureido groups.

Isothiocyanate derivatives are useful and widely used as building blocks in the synthesis of nitogen, sulfur and oxygen containing heterocyclic compounds and organometallic compounds of academic, pharmaceutical and industrial interest. Thus, the isothiocyanate derivative **18** was synthesized by treatment of compound **1** with ammonium thiocyanate in dry acetone for 1 h. This method lead to a higher overall yield and shorter working time in comparison with the reported method by using silver thiocyanate.[23] IR spectrum of **18** showed band at 2072 cm^{-1} (N=C=S). The reactivity of isothiocyanate derivative **18** towards amino compounds such as sulfonamides was discussed. Thus, interaction of compound **18** with sulfonamides in DMF in the presence of triethylamine yielded the corresponding carbamimidothioic acid derivatives **19**–**25**, respectively. The structures of compounds **19**–**25** were supported by analytical and spectral data. The IR spectra of compounds **19**–**25** showed the absence of (N=C=S) band and presence of bands for (NH), (C=N), (SO_2) and (C–Cl) groups. ^{1}H-NMR spectra in (DMSO-d_6) for compounds **19**–**25** showed signals at 2.2–2.5 ppm for SH groups, and 8.4–8.9 ppm due to SO_2NH group.

Molecular Docking Oncology drug discovery has benefited significantly from progress in understanding how to target kinases with small molecules relative to other disease indications. The constitutive activity of this class of kinase target makes them essential for survival and/or proliferation of the cancer cell. This so-called oncogene addiction.[24,25] A recent example is that PTen-deficient cancers depend on PI3Kβ to sustain activation of the PI3K pathway, whereas PI3Kα kinase activity appears to be required to sustain the proliferation of established tumours.[26,27] Like the majority of protein kinase inhibitors, all existing PI3K inhibitors bind competitively in the ATP-binding pocket of the catalytic domain. This strategy has enabled the development of both pan-PI3K- and isoform-specific inhibitors. Loosely discriminate inhibitors that target multiple PI3K isoforms may more thoroughly shut down PI3K signaling for the treatment of acute life-threatening diseases.[28]

The PI3K (phosphoinositide 3-kinase) pathway is often overactive in human cancers, and various genetic alterations have been found to cause this. In all cases, PI3K inhibition is considered to be one of the most promising targeted therapies for cancer treatment. In our investigation three different cell lines were chosen for *in vitro* assay. It was reported that these cell lines were affected by PI3K inhibitors with high cytotoxic

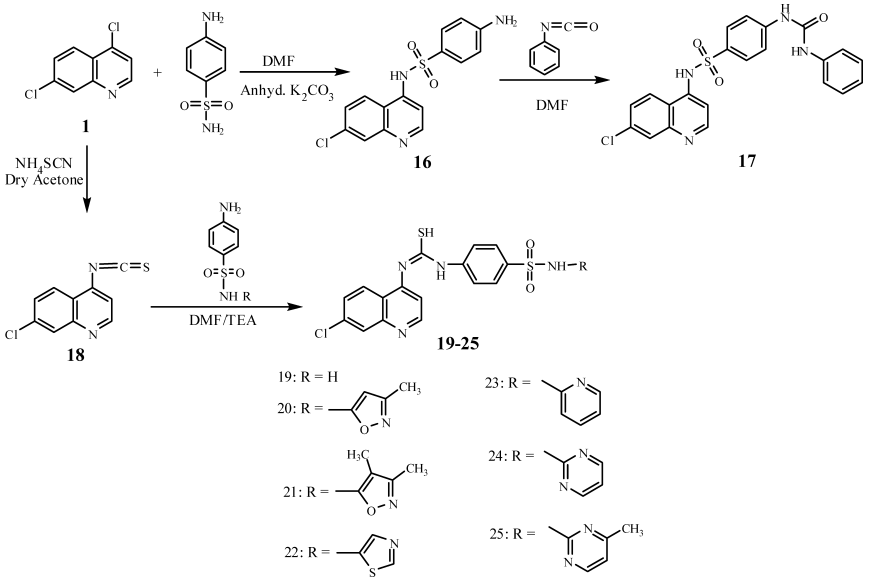

Chart 3. Synthetic Pathways for Compounds **16**–**25**

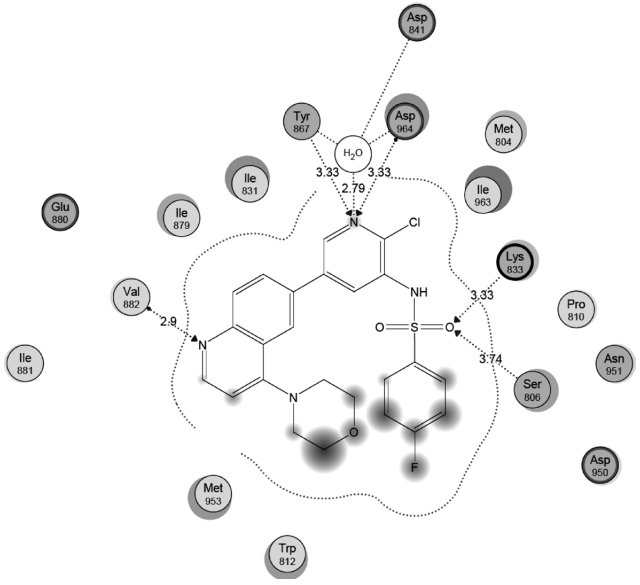

Fig. 1. Co-crystallized Quinoline Ligand on the Active Site of Phosphoinositide Kinase (PI3K)

activity.[29,30]

Based on the previous findings and as a trial to suggest the mechanism of action of the cytotoxic activity for the synthesized compounds docking of all newly synthesized compounds was done on the active site of PI3K.

The protein data bank file (PDB: 3S2A) was selected for this purpose. The file contains PI3K enzyme co-crystallized with a quinoline ligand. All docking procedures were achieved by Molecular Operating Environment (MOE) software 10.2008 provided by chemical computing group, Canada. Docking on the active site of PI3K enzyme was performed for all synthesized compounds.

Docking protocol was verified by redocking of the co-crystallized ligand in the vicinity of the active site of the enzyme with energy score (S)=−29.8249 kcal/mol and root mean standard deviation (RMSD)=1.9094, Fig. 1. The quinoline ligand interacts with the active site of PI3K by six interactions: Val 882 with a hydrogen bond of 2.90 Å, Tyr 867 with a hydrogen bond of 3.33 Å, Asp 864 with a hydrogen bond of 3.33 Å, Lys 833 with a hydrogen bond of 3.33 Å, Ser 806 with a hydrogen bond of 3.74 Å and Asp 841 with a hydrogen bond of 2.79 Å through a water molecule. All synthesized compounds were fit to the active site of PI3K enzyme with good energy scores (S) suggesting activity as PI3K inhibitors. Energy scores (S) and amino acid interactions for the synthesized compounds were listed in Table 1. Compound **21** gave the best energy score (S)=−24.4292 and interacted with Lys 802 with a hydrogen bond of 3.76 Å, Lys 833 with a hydrogen bond of 2.64 Å, Ser 806 with a hydrogen bond of 2.33 Å, Lys 805 with a hydrogen bond of 2.88 Å and with His 867 with a hydrogen bond of 3.83 Å, Fig. 2.

The best amino acid interactions were formed with compound **19** with Lys 890 with a hydrogen bond of 2.92 Å, Lys 833 with a hydrogen bond of 2.52, 3.06 Å, Asp 864 with a hydrogen bond of 3.24, 3.62 Å and Tyr 867 through a water molecule with a hydrogen bond of 2.93 Å, Fig. 3.

***In Vitro* Antitumor Activity** The newly synthesized compounds were evaluated for their *in vitro* cytotoxic activity against human breast cancer cell line (MDA-MB231), skin cancer cell line (HT 1080) and neuroblastoma cell line (SH-SY5Y). Doxorubicin which is one of the most effective anticancer agents was used as the reference drug in this study. The relationship between surviving fraction and drug concentration was plotted to obtain the survival curve of cancer cell lines. The response parameter calculated was the IC_{50} value, which corresponds to the concentration required for 50% inhibition of cell viability. Table 2 shows the *in vitro* cytotoxic activity of the synthesized compounds where all compounds exhibited moderate activity compared to the reference drug in case of human breast cancer cell line (MDA-MB231), while compounds **19**–**25** where inactive in case of skin cancer cell line (HT 1080). Also compounds **20**–**23** and **25** were inactive in case of neuroblastoma cell line (SH-SY5Y).

All the synthesized compounds showed moderate cytotoxic activity compared to Doxorubicin on human breast cancer cell line (MDA-MB231). The

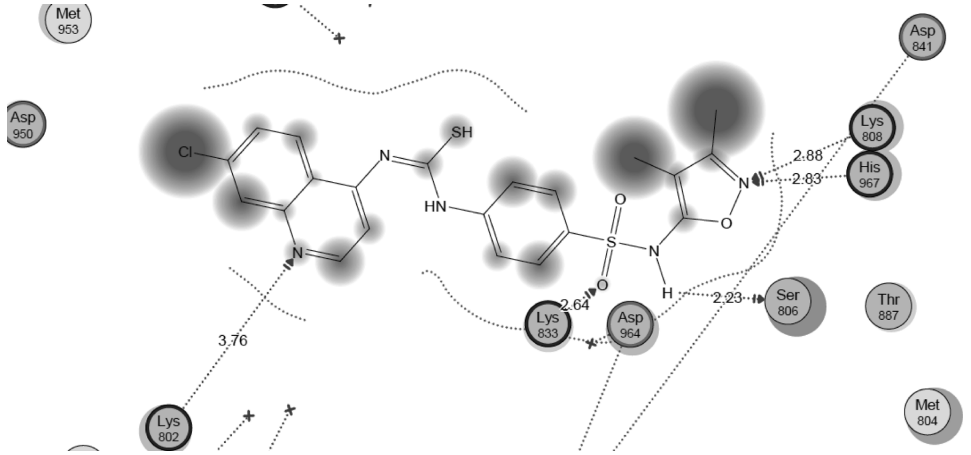

Fig. 2. Compound **21** on the Active Site of PI3K

Table 1. Binding Scores and Amino Acid Interactions of the Docked Compounds on the Active Site of Phosphoinisitol Kinase (PI3K)

Compound No.	S (kcal/mol)	Amino acid interactions	Interacting groups	H bond length Å
2	−12.3409	Lys 833, Asp 841 (water) Val 882, Asp 964	SO_2, *N*-quinoline, SO_2, SO_2	3.02, 2.543.42, 3.63
3	−17.9656	Lys 833, Asp 841 (water) Lys 890, Asp 964	SO_2, NHCO, *N*-quinoline, NHCO	3.31, 2.672.59, 3.33
4	−16.2910	Lys 890, Tyr 867 Asp 864, Asp 841	*N*-quinoline, SO_2SO_2, SO_2	2.82, 3.642.85, 2.66
5	−21.0847	Lys 833, Lys 890	SO_2, *N*-quinoline	2.73, 2.50
6	−20.0851	Lys 890, Lys 833 Ser 806, Asp 864	*N*-quinoline, SO_2SO_2, NH	2.61, 2.353.28, 1.25
7	−22.2882	Lys 890, Asp 846 Tyr 867, Lys 833	*N*-quinoline, SO_2SO_2, *N*-thiazole	2.91, 3.803.33, 2.31
8	−20.8781	Lys 833, Lys 890 Lys 890	*N*-quinoline, SO_2SO_2	2.67, 2.52–2.90
9	−21.7370	Asp 864 (water), Lys 890	*N*-quinoline, SO_2	3.72, 2.38
10	−20.0596	Val 882, Lys 802 Lys 833	*N*-quinoline, SO_2SO_2	3.25, 2.433.41
11	−19.5996	Lys 833, Asp 841 (water)	*N*-quinoline, SO_2	3.20, 3.56
12	−22.4876	Asp 864 (water), Tyr 867 Lys 890, Lys 890	*N*-quinoline, SO_2, *N*-pyrimidine	3.02, 3.763.35, 2.50
13	−21.3247	Tyr 867, Asp 864 (water) Lys 890, Lys 890	*N*-quinoline, SO_2	3.17, 2.723.66, 3.86
14	−21.2363	Asp 841 (water), Lys 890 Lys 802	*N*-quinoline, SO_2OCH_3	3.55, 2.453.03
15	−21.8738	Val 882, Ser 806 Lys 833, Asp 864	*N*-quinoline, $NHSO_2$, NH	3.56, 2.972.81, 2.92
16	−14.8566	Val 882, Tyr 864 Asp 864 (water), Asp 864	*N*-quinoline, SO_2SO_2, NH_2	3.05, 3.093.21, 1.39
17	−19.3479	Ser 806, Asp 864 Tyr 867, Asp 864 (water)	*N*-quinoline, SO_2SO_2	2.82, 3.013.78, 3.60
19	−20.3214	Lys 890, Lys 833 Lys 833, Asp 864Asp 864, Tyr 867 (water)	*N*-quinoline, $NHSO_2$, $NHSO_2$	2.92, 3.523.60, 3.243.62, 2.93
20	−18.4624	Lys 890, Ser 806 Lys 833, Asp 864	*N*-quinoline, SO_2SO_2, SO_2NH	2.82, 3.593.37, 1.48
21	−24.4292	Lys 802, Lys 833 Ser 806, Lys 805	*N*-quinoline, SO_2SO_2NH, *N*-isoxazole	3.76, 2.642.33, 2.88
22	−17.4305	Val 882, Lys 833 Ser 806, Asp 864	*N*-quinoline, SO_2SO_2, SO_2NH	3.60, 2.743.21, 1.24
23	−20.9130	Lys 890, Tyr 867 Asp 864, Asp 841 (water)	*N*-quinoline, SO_2SO_2	2.63, 3.183.06, 3.41
24	−23.4308	Asp 841 (water), Lys 833 Lys 833, Lys 805	*N*-quinoline, SO_2SO_2, imino	2.80, 2.282.95, 3.00
25	−23.3010	Asp 841 (water), Lys 833 Asp 864, Lys 805	*N*-quinoline, imino, NH, SO_2	3.29, 2.342.54, 3.27

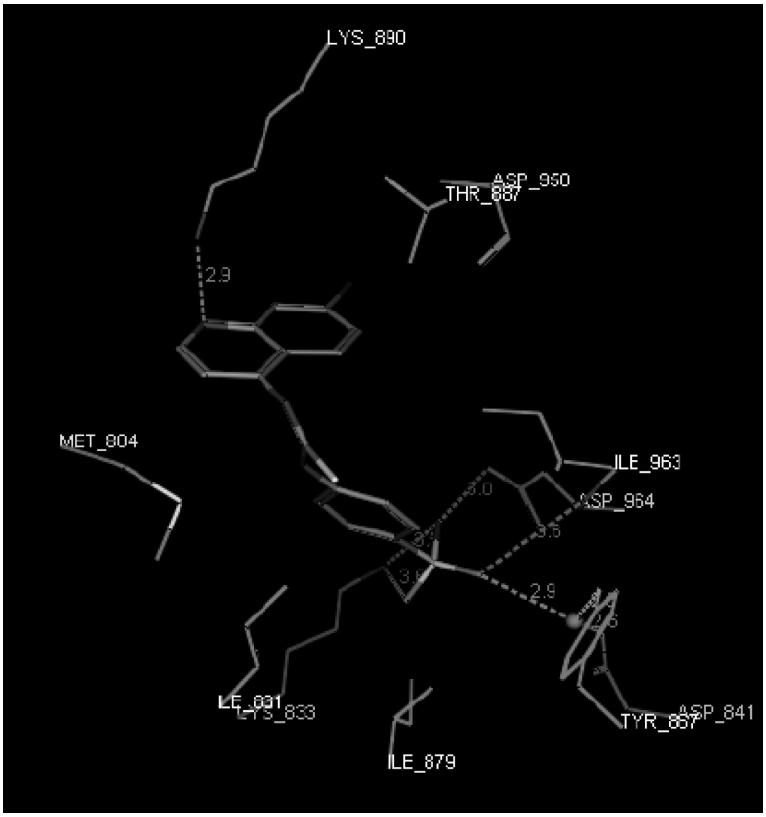

Fig. 3. Compound **19** on the Active Site of PI3K

Table 2. *In Vitro* Anticancer Screening of the Synthesized Compounds against Human Breast Cells, Skin Cancer Cells and Neuroblastoma

Compound No.	MDA-MB231 (breast cancer cells)	HT 1080 (skin cancer cells)	SH-SYSY (neuroblastoma cells)
	IC_{50} (μM)		
2	114.40	173.50	43.92
3	154.09	223.24	50.42
4	129.55	230.82	59.12
5	135.01	156.07	115.46
6	82.88	219.99	106.41
7	137.73	139.72	85.14
8	154.87	177.22	56.01
9	117.43	136.41	76.64
10	112.70	139.42	130.46
11	115.46	128.55	105.90
12	103.53	94.66	87.40
13	97.47	127.35	204.70
14	95.43	107.22	106.79
15	70.21	109.40	84.23
16	107.74	140.99	72.64
17	64.41	75.05	30.71
19	45.92	NA	35.41
20	88.05	NA	NA
21	120.99	NA	NA
22	97.41	NA	NA
23	72.09	NA	NA
24	68.66	NA	124.77
25	78.50	NA	NA
Doxorubicin	82.53	88.32	73.72

N-(4-sulfamoylphenyl)carbamimidothioic acid derivative **19** was the most active compound with IC_{50} value 45.92 μM which was better than that of Doxorubicin. On the other hand, quinoline sulfonamide derivatives **2**–**14** showed IC_{50} values ranging from 82.88–154.87 μM. The unsubstituted sulfonamide derivative **2** showed IC_{50} value 114.4.40 μM. Upon substitution on N of sulfonamide group with acetyl group as in compound **3** or guanidine as in compound **4** or 5-membered heterocyclic substituents as in compounds **5**–**8** the activity drops except for compound **6** with dimethyl isoxazole substituent in which the activity increased with IC_{50} value 82.88 μM. This was not the case in 6-membered substitutions as in compounds **9**–**14** as the activity tends to be the same or increased especially with 5,6-dimethoxypyrimidine substituent as in compound **14** with IC_{50} value 95.43 μM. Compounds **16**–**25** showed better activity except for compound **21** with IC_{50} value 120.99 μM. The best two compounds were the unsubstituted sulfonamide urea derivative **17** with IC_{50} value 64.41 μM and the unsubstituted cabamimidothioic acid derivative **19** with IC_{50} value 45.92 μM.

Most of the synthesized compounds showed moderate cytotoxic activity on skin cancer cell line (HT 1080) except for compounds **19**–**25**. The most active compound was the unsubstituted sulfonamide derivative **17** with IC_{50} value 75.05 μM. For compounds **2**–**14** better activity also was observed for 6-membered substituted sulfonamide derivatives **9**–**14** than unsubstituted **2**, acetyl **3**, guanidine **4** and 5-membered substituted derivatives **5**–**8**. On the other hand, the sulfonamide phenyl urea derivative **15** showed better activity than compound **16** with IC_{50} value 109.40 μM and 140.99 μM, respectively.

Most of the synthesized compounds showed moderate cytotoxic activity compared to Doxorubicin on neuroblastoma cell

line (SH-SY5Y) except for compounds **20**–**23** and **25**. Activity on this cell line was much higher for all active compounds with IC_{50} values in the range 30.71–130.46 μM. The most active compound was compound **17** with IC_{50} value 30.71 μM. Also a remarkable activity was attributed to compound **19** with IC_{50} value 35.41 μM. Compounds **2**–**16** showed IC_{50} values 43.92–204.70 μM while compound **24** showed IC_{50} value 124.77 μM.

Relationship between Molecular Docking and *in Vitro* Screening The inhibition of PI3K could be a suggested mechanism for the cytotoxic activity of the synthesized compound. This can be demonstrated by the docking results that were performed on the active site of PI3K. The co-crystalized quinoline ligand intearcts with the amino acids of the active site by six interaction that were mentioned before. On the other hand, the newly synthesized compounds interact with these amino acids by 1, 2, 3 or 4 interactions. We could postulate a relationship between the number on amino acid interactions and the *in vitro* cytotoxic activity on the used cell lines. Most of the synthesized compounds interact with the amino acids of the active site by 3 interactions. Compounds **2**, **3**, **4**, **6**, **7**, **16**, **17**, **19**, **20**, **23** and **25** showing good activity on one or more cell lines with IC_{50} values 43.92 μM (neuroblastoma), 50.42 μM (neuroblastoma), 59.12 μM (neuroblastoma), 82.88 μM (MDA-MB231), 85.14 μM (neuroblastoma), 72.64 μM (neuroblastoma), 30.71 μM (neuroblastoma), 35.41 μM (neuroblastoma), 88.05 μM (MDA-MB321), 72.09 μM (MDA-MB321) and 78.50 μM (MDA-MB321), respectively. Compounds **15** and **22** interact with the amino acids of the active site by 4 interactions showing good cytotoxic activity for compound **15** and moderate activity for compound **22** with IC_{50} values of 70.21 μM (MD-MB321) and 97.41 μM (MD-MB321), respectively. This could be explained by the weak docking score of compound **22** (−17.4305 kcal/mol) indicating improper fitting on the active site of the enzyme PI3K. The rest on the newly synthesized compounds interact with amino acids of the active site by one or two interactions showing moderate, weak or no activity in the *in vitro* screening.

Experimental

Chemistry Melting points (°C, uncorrected) were determined in open capillaries on a Gallenkemp melting point apparatus (Sanyo Gallenkemp, Southborough, U.K.) and were uncorrected. Pre-coated silica gel plates (silica gel 0.25 mm, 60 G F254; Merck, Germany) were used for thin layer chromatography, dichloromethane–methanol (9.5 : 0.5) mixture was used as a developing solvent system and the spots were visualized by UV light and/or iodine. Infra-red spectra were recorded in KBr discs using IR-470 Shimadzu spectrometer (Shimadzu, Tokyo, Japan). NMR spectra (in DMSO-d_6) were recorded on Bruker AC-300 Ultra Shield NMR spectrometer (Bruker, Flawil, Switzerland, δ ppm) at 500 MHz using TMS as internal standard and peak multiplicities are designed as follows: s, singlet; d, doublet; t, triplet; m, multiplet. Elemental analyses were performed on Carlo Erba 1108 Elemental Analyzer (Heraeus, Hanau, Germany).

General Procedure for the Synthesis of Compounds 2–14 A mixture of **1** (1.98 g, 0.01 mol) and the corresponding sulfonamide (0.012 mol) in dry DMF (20 mL) was refluxed for 6 h. The solid obtained after concentration was filtered and crystallized from dioxane to give **2**–**14**.

4-(7-Chloroquinolin-4-yl-amino)benzenesulfonamide (**2**): Yield 94%, melting point 263.4°C. IR: ν_{max}/cm^{-1} 3420, 3296 (NH, NH_2), 1610 (C=N), 1325, 1155 (SO_2), 817 (C–Cl). ^{1}H-NMR (DMSO-d_6, D_2O) δ: 6.9, 8.5 (2d, 2H, 2CH pyridine, J=7.4 Hz), 7.1–7.9 (m, 9H, Ar-H+SO_2NH_2), 10.7 (s, 1H, NH, D_2O-exchangeable). ^{13}C-NMR (DMSO-d_6) δ: 112.4, 118.7 (2), 121.2, 125.4, 126.7, 127.2 (2), 127.3, 135.2, 139.8, 146.8, 148.4, 149.8, 151.9. *Anal.* Calcd for $C_{15}H_{12}ClN_3O_2S$ (333.79): C, 53.97; H, 3.62; N, 12.59. Found: C, 53.76; H, 3.82; N, 12.33.

N-(4-(7-Chloroquinolin-4-yl-amino)phenylsulfonyl)-acetamide (**3**): Yield 88%, melting point 245.5°C. IR: ν_{max}/cm^{-1} 3489, 3320 (NH), 3053 (CH arom.), 2920, 2899 (CH aliph.), 1650 (C=O), 1614 (C=N), 1360, 1165 (SO_2), 815 (C–Cl). ^{1}H-NMR (DMSD-d_6, D_2O) δ: 3.1 (s, 3H, $COCH_3$), 6.9, 8.6 (2d, 2H, 2CH pyridine, J=7.8 Hz), 7.0–8.3 (m, 7H, Ar-H), 11.4 (s, 1H, NHPh, D_2O-exchangeable), 15.1 (s, 1H, NHCO, D_2O-exchangeable). ^{13}C-NMR (DMSD-d_6) δ: 19.2, 114.2, 116.3(2), 119.3, 125.0, 126.3, 127.3(2), 127.5, 138.4, 139.1, 140.2, 142.2, 143.7, 154.3, 176.4. *Anal.* Calcd for $C_{17}H_{14}ClN_3O_3S$ (375.83): C, 54.33; H, 3.75; N, 11.18. Found: C, 54.60; H, 3.51; N, 11.38.

N-Carbamimidoyl-4-(7-chloroquinolin-4-yl-amino)-benzenesulfonamide (**4**): Yield 84%, melting point 137.1°C. IR: ν_{max}/cm^{-1} 3470, 3300 (NH, NH_2), 3075 (CH arom.), 1612 (C=N), 1325, 1134 (SO_2), 841 (C–Cl). ^{1}H-NMR (DMSD-d_6, D_2O) δ: 4.7 (hump, 1H, NH imino, D_2O-exchangeable), 6.6, 8.5 (2d, 2H, 2CH pyridine, J=7.5 Hz), 6.9 (s, 2H, NH_2, D_2O-exchangeable), 7.0–8.4 (m, 8H, Ar-H+SO_2NH), 10.4 (s, 1H, NHPh, D_2O-exchangeable). ^{13}C-NMR (DMSD-d_6) δ: 114.9, 117.3(2), 122.3, 123.2, 125.1, 126.2(2), 127.1, 136.2, 140.4, 144.1, 147.7, 150.1, 152.7, 158.1. *Anal.* Calcd for $C_{16}H_{14}ClN_5O_2S$ (375.83): C, 51.13; H, 3.75; N, 18.63. Found: C, 51.41; H, 3.48; N, 18.94.

4-(7-Chloroquinolin-4-yl-amino)-*N*-(3-methyl-isoxazol-5-yl)benzenesulfonamide (**5**): Yield 91%, melting point 162.9°C. IR: ν_{max}/cm^{-1} 3320, 3210 (NH), 3059 (CH arom.), 2926, 2812 (CH aliph.), 1616 (C=N), 1375, 1145 (SO_2), 813 (C–Cl). ^{1}H-NMR (DMSD-d_6, D_2O) δ: 2.3 (s, 3H, CH_3), 6.7 (s, 1H, CH isoxazole), 6.8, 8.5 (2d, 2H, 2CH pyridine, J=7.2 Hz), 7.1–8.4 (m, 8H, Ar-H+SO_2NH), 10.6 (s, 1H, NHPh, D_2O-exchangeable). ^{13}C-NMR (DMSD-d_6) δ: 12.0, 103.1, 115.2, 118.6(2), 121.5, 123.3, 127.2, 127.7(2), 128.7, 131.2, 139.1, 146.1, 148.8, 149.2, 152.8, 157.6, 162.2. *Anal.* Calcd for $C_{19}H_{15}ClN_4O_3S$ (414.87): C, 55.01; H, 3.64; N, 13.50. Found: C, 55.34; H, 3.91; N, 13.76.

4-(7-Chloroquinolin-4-yl-amino)-*N*-(3,4-dimethyl-isoxazol-5-yl)benzenesulfonamide (**6**): Yield 88%, melting point 169.0°C. IR: ν_{max}/cm^{-1} 3380, 3253 (NH), 3057 (CH arom.), 2924, 2840 (CH aliph.), 1609 (C=N), 1375, 1155 (SO_2), 815 (C–Cl). ^{1}H-NMR (DMSD-d_6, D_2O) δ: 2.2, 2.3 (2s, 6H, $2CH_3$), 7.1, 8.5 (2d, 2H, 2CH pyridine, J=8.1 Hz), 7.2–8.3 (m, 8H, Ar-H+SO_2NH), 10.6 (s, 1H, NHPh, D_2O-exchangeable). ^{13}C-NMR (DMSD-d_6) δ: 10.8, 14.6, 103.1, 115.7, 118.2(2), 121.3, 125.3, 125.9, 127.2(2), 127.5, 135.3, 138.9, 143.0, 146.6, 148.5, 149.7, 159.6, 162.2. *Anal.* Calcd for $C_{20}H_{17}ClN_4O_3S$ (428.89): C, 56.1; H, 4.00; N, 13.06. Found: C, 55.77; H, 3.68; N, 13.36.

4-(7-Chloroquinolin-4-yl-amino)-*N*-(thiazol-5-yl)-benzenesulfonamide (**7**): Yield 90%, melting point 181.6°C. IR: ν_{max}/cm^{-1} 3410, 3270 (NH), 3097 (CH arom.), 1616 (C=N), 1375, 1141 (SO_2), 815 (C–Cl). ^{1}H-NMR (DMSD-d_6, D_2O) δ: 6.7, 8.6 (2d, 2H, 2CH pyridine, J=7.7 Hz), 6.9–8.3 (m,

10H, Ar-H+SO_2NH), 10.6 (s, 1H, NHPh, D_2O-exchangeable). ^{13}C-NMR (DMSD-d_6) δ: 115.2, 117.4(2), 118.7, 123.3, 126.3, 127.3(2), 127.6, 131.2, 137.8, 144.6, 148.1, 149.8, 152.8, 160.0, 162.2, 168.6. *Anal.* Calcd for $C_{18}H_{13}ClN_4O_2S_2$ (416.90): C, 51.86; H, 3.14; N, 13.44. Found: C, 51.59; H, 3.52; N, 13.12.

4-(7-Chloroquinolin-4-yl-amino)-*N*-(1-phenyl-1*H*-pyrazol-5-yl)benzenesulfonamide (**8**): Yield 81%, melting point 159.0°C. IR: ν_{max}/cm^{-1} 3348, 3244 (NH), 3064 (CH arom.), 1615 (C=N), 1375, 1155 (SO_2), 817 (C–Cl). ^{1}H-NMR (DMSD-d_6, D_2O) δ: 6.6, 8.4 (2d, 2H, 2CH pyridine, J=7.7 Hz), 6.8–8.2 (m, 15H, Ar-H+SO_2NH), 10.5 (s, 1H, NHPh, D_2O-exchangeable). ^{13}C-NMR (DMSD-d_6) δ: 97.6, 112.5, 118.8(2), 119.0, 122.8, 125.2, 127.3, 127.9, 128.3(2), 128.8, 133.6, 134.4, 135.3, 136.5, 139.5, 141.7, 144.7, 146.5, 148.9, 151.5, 153.1. *Anal.* Calcd for $C_{24}H_{18}ClN_5O_2S$ (475.95): C, 60.56; H, 3.81; N, 14.71. Found: C, 60.22; H, 3.57; N, 14.44.

4-(7-Chloroquinolin-4-yl-amino)-*N*-(pyridin-2-yl)-benzenesulfonamide (**9**): Yield 87%, melting point 205.6°C. IR: ν_{max}/cm^{-1} 3420, 3232 (NH), 3057 (CH arom.), 1635 (C=N), 1394, 1138 (SO_2), 773 (C–Cl). ^{1}H-NMR (DMSD-d_6, D_2O) δ: 6.9, 8.6 (2d, 2H, 2CH pyridine, J=7.8 Hz), 7.0–8.3 (m, 11H, Ar-H), 10.6 (s, 1H, NHPh, D_2O-exchangeable), 12.1 (s, 1H, SO_2NH, D_2O-exchangeable). ^{13}C-NMR (DMSD-d_6) δ: 108.2, 114.0, 115.5, 117.4(2), 118.7, 125.2, 127.8, 128.1(2), 128.2, 128.8, 136.6, 142.5, 143.2, 146.0(2), 148.7, 149.2, 153.1. *Anal.* Calcd for $C_{20}H_{15}ClN_4O_2S$ (410.88): C, 58.46; H, 3.68; N, 13.64. Found: C, 58.77; H, 3.46; N, 13.96.

4-(7-Chloroquinolin-4-yl-amino)-*N*-(pyrimidin-2-yl)-benzenesulfonamide (**10**): Yield 89%, melting point 174.6°C. IR: ν_{max}/cm^{-1} 3448, 3390 (NH), 3057 (CH arom.), 1637 (C=N), 1376, 1159 (SO_2), 812 (C–Cl). ^{1}H-NMR (DMSD-d_6, D_2O) δ: 6.6, 8.6 (2d, 2H, 2CH pyridine, J=7.3 Hz), 7.0–8.5 (m, 10H, Ar-H), 10.6 (s, 1H, NHPh, D_2O-exchangeable), 11.9 (s, 1H, SO_2NH, D_2O-exchangeable). ^{13}C-NMR (DMSD-d_6) δ: 108.4, 112.1, 118.6(2), 121.8, 123.7, 127.1, 128.9(2), 129.2, 129.7, 135.1, 144.5, 147.6, 150.3, 153.0, 158.3(2), 162.2. *Anal.* Calcd for $C_{19}H_{14}ClN_5O_2S$ (411.86): C, 55.41; H, 3.43; N, 17.00. Found: C, 55.71; H, 3.11; N, 17.37.

4-(7-Chloroquinolin-4-yl-amino)-*N*-(4-methyl-pyrimidin-2-yl)benzenesulfonamide (**11**): Yield 87%, melting point 164.0°C. IR: ν_{max}/cm^{-1} 3415, 3310 (NH), 3080 (CH arom.), 2940, 2866 (CH aliph.), 1618 (C=N), 1340, 1157 (SO_2), 817 (C–Cl). ^{1}H-NMR (DMSD-d_6, D_2O) δ: 2.2 (s, 3H, CH_3), 6.6, 8.6 (2d, 2H, 2CH pyridine, J=7.4 Hz), 7.0–8.3 (m, 9H, Ar-H), 10.5 (s, 1H, NHPh, D_2O-exchangeable), 11.8 (s, 1H, SO_2NH, D_2O-exchangeable). ^{13}C-NMR (DMSD-d_6) δ: 23.3, 111.9, 115.3, 118.3(2), 120.7, 124.7, 127.2, 129.1(2), 129.5, 130.0, 135.0, 145.8, 148.8, 149.1, 152.9, 157.4, 162.2, 168.2. *Anal.* Calcd for $C_{20}H_{16}ClN_5O_2S$ (425.89): C, 56.40; H, 3.79; N, 16.44. Found: C, 56.12; H, 3.49; N, 16.16.

4-(7-Chloroquinolin-4-yl-amino)-*N*-(4,6-dimethyl-pyrimidin-2-yl)benzenesulfonamide (**12**): Yield 79%, melting point 176.7°C. IR: ν_{max}/cm^{-1} 3448, 3395 (NH), 3086 (CH arom.), 2924, 2861 (CH aliph.), 1597 (C=N), 1345, 1140 (SO_2), 866 (C–Cl). ^{1}H-NMR (DMSD-d_6, D_2O) δ: 2.2 (s, 6H, $2CH_3$), 6.6, 8.6 (2d, 2H, 2CH pyridine, J=7.3 Hz), 7.0–8.5 (m, 8H, Ar-H), 10.3 (s, 1H, NHPh, D_2O-exchangeable), 11.9 (s, 1H, SO_2NH, D_2O-exchangeable). ^{13}C-NMR (DMSD-d_6) δ: 22.8, 23.0, 104.0, 113.4, 117.4(2), 119.8, 125.9, 127.2, 129.2, 129.3(2), 129.8, 130.2, 135.0, 144.1, 147.6, 150.3, 156.3, 162.2(2), 167.3. *Anal.* Calcd for $C_{21}H_{18}ClN_5O_2S$ (439.92): C, 57.33; H, 4.12; N, 15.92. Found: C, 57.60; H, 4.41; N, 15.62.

4-(7-Chloroquinolin-4-yl-amino)-*N*-(2,6-dimethoxy-pyrimidin-4-yl)benzenesulfonamide (**13**): Yield 82%, melting point 264.7°C. IR: ν_{max}/cm^{-1} 3370, 3248 (NH), 3066 (CH arom.), 2930, 2812 (CH aliph.), 1610 (C=N), 1381, 1134 (SO_2), 823 (C–Cl). ^{1}H-NMR (DMSD-d_6, D_2O) δ: 3.7 (s, 6H, $2OCH_3$), 5.9 (s, 1H, CH pyrimidine), 6.6, 8.6 (2d, 2H, 2CH pyridine, J=7.5 Hz), 7.0–8.3 (m, 7H, Ar-H), 10.2 (s, 1H, NHPh, D_2O-exchangeable), 10.7 (s, 1H, SO_2NH, D_2O-exchangeable). ^{13}C-NMR (DMSD-d_6) δ: 53.6, 54.3, 84.1, 112.4, 118.6(2), 121.0, 123.8, 127.2, 129.0(2), 129.3, 133.0, 137.2, 147.8, 148.6, 150.5, 153.3, 162.2, 163.3, 173.6. *Anal.* Calcd for $C_{21}H_{18}ClN_5O_4S$ (471.92): C, 53.45; H, 3.84; N, 14.84. Found: C, 53.21; H, 3.63; N, 14.66.

4-(7-Chloroquinolin-4-yl-amino)-*N*-(5,6-dimethoxy-pyrimidin-4-yl)benzenesulfonamide (**14**): Yield 77%, melting point 183.6°C. IR: ν_{max}/cm^{-1} 3340, 3248 (NH), 3070 (CH arom.), 2940, 2831 (CH aliph.), 1375, 1159 (SO_2), 821 (C–Cl). ^{1}H-NMR (DMSD-d_6, D_2O) δ: 3.7 (s, 6H, $2OCH_3$), 6.6, 8.4 (2d, 2H, 2CH pyridine, J=7.2 Hz), 7.0–8.3 (m, 8H, Ar-H), 9.5 (s, 1H, NHPh, D_2O-exchangeable), 12.3 (s, 1H, SO_2NH, D_2O-exchangeable). ^{13}C-NMR (DMSD-d_6) δ: 58.9(2), 113.9, 119.2(2), 120.0, 123.4, 127.6, 129.0(2), 129.2, 129.7, 133.6, 134.2, 145.1, 146.9, 149.4, 150.6, 151.8, 157.9, 161.4. *Anal.* Calcd for $C_{21}H_{18}ClN_5O_4S$ (471.92): C, 53.67; H, 3.84; N, 14.84. Found: C, 53.45; H, 3.51; N, 14.49.

4-(7-Chloroquinolin-4-yl-amino)-*N*-(phenylcarbamoyl)-benzenesulfonamide (15) A mixture of **2** (3.3 g, 0.01 mol) and phenyl isocyanate (1.21 g, 0.01 mol) in dry DMF (20 mL) containing anhydrous K_2CO_3 (1 g) was refluxed for 14 h. The obtained solid was filtered and crystallized from dioxane. Yield 88%, melting point 159.8°C, IR: ν_{max}/cm^{-1} 3340, 3280, 3186 (NH), 3055 (CH arom.), 1660 (C=O), 1620 (C=N), 1373, 1172 (SO_2), 744 (C–Cl). ^{1}H-NMR (DMSD-d_6, D_2O) δ: 6.6, 8.2 (2d, 2H, 2CH pyridine, J=7.1 Hz), 7.0–8.3 (m, 12 H, Ar-H), 8.4 (s, 1H, NHCO, D_2O exchangeable), 10.2 (s, 1H, NHPh, D_2O-exchangeable) 12.8 (s, 1H, SO_2NH, D_2O exchangeable). ^{13}C-NMR (DMSD-d_6) δ: 112.4, 117.2(2), 117.7, 121.0 (2), 121.7, 125.4, 127.3, 127.9 (2), 128.7 (2), 129.0, 129.6, 136.7, 139.0, 140.3, 141.3, 142.9, 151.7, 159.6. *Anal.* Calcd for $C_{22}H_{17}ClN_4O_3S$ (452.91): C, 58.34; H, 3.78; N, 12.37. Found: C, 58.70; H, 3.49; N, 12.61.

4-Amino-*N*-(7-chloroquinolin-4-yl)benzenesulfonamide (16) A mixture of **1** (1.98 g, 0.01 mol) and sulfanilamide (1.72 g, 0.01 mol) in dry DMF (20 mL) containing anhydrous K_2CO_3 (1 g) was refluxed for 6 h. The reaction mixture was poured onto ice/water and the obtained solid was crystallized from ethanol. Yield 94%, melting point 155.9°C, IR: ν_{max}/cm^{-1} 3454, 3389, 3244 (NH, NH_2), 3050 (CH arom.), 1627 (C=N), 1363, 1136 (SO_2), 808 (C–Cl). ^{1}H-NMR (DMSD-d_6, D_2O) δ: 5.7 (s, 2H, NH_2, D_2O exchangeable), 6.5, 8.6 (2d, 2H, 2CH pyridine, J=7.0 Hz), 7.0–8.3 (m, 8 H, Ar-H+SO_2NH). ^{13}C-NMR (DMSD-d_6) δ: 112.5, 117.8 (2), 119.6, 124.7, 127.2, 128.6 (2), 129.0, 134.0, 136.2, 147.9, 149.3, 151.7, 153.0. *Anal.* Calcd for $C_{15}H_{12}ClN_3O_2S$ (333.79): C, 53.97; H, 3.62; N, 12.59. Found: C, 53.68; H, 3.41; N, 12.31.

***N*-(7-Chloroquinolin-4-yl)-4-(3-phenylureido)-benzenesulfonamide (17)** A mixture of **16** (3.33 g, 0.01 mol) and phenyl isocyanate (1.21 g, 0.01 mol) in dry DMF (20 mL) was heated under reflux for 10 h. The obtained solid was crystallized from DMF. Yield 79%, melting point 194.0°C,

IR: ν_{max}/cm^{-1} 3375, 3224 (NH), 3066 (CH arom.), 1678 (C=O), 1618 (C=N), 1376, 1132 (SO_2), 808 (C–Cl). ^{1}H-NMR (DMSD-d_6, D_2O) δ: 6.9, 8.4 (2d, 2H, 2CH pyridine, J=7.2 Hz), 7.0–8.3 (m, 13 H, Ar-H+SO_2NH), 9.1, 9.2 (2s, 2H, 2NH, D_2O exchangeable). ^{13}C-NMR (DMSD-d_6) δ: 117.3, 120.0, 121.7 (2), 122.8, 124.5 (2), 126.7, 127.5, 128.7 (2), 129.0 (2), 129.4, 134.2, 139.3, 139.8, 148.4, 151.3, 151.8, 152.5, 162.2. *Anal.* Calcd for $C_{22}H_{17}ClN_4O_3S$ (452.91): C, 58.34; H, 3.78; N, 12.37. Found: C, 58.72; H, 3.55; N, 12.18.

7-Chloro-4-isothiocyanatoquinoline (18) A mixture of **1** (1.98 g, 0.01 mol) and ammonium thiocyanate (1.52 g, 0.02 mol) in dry acetone (20 mL) was refluxed for 1 h. The reaction mixture was cooled, poured onto ice/water and the obtained solid was crystallized from ethanol to give **18** (mp and mpp as reported). Yield 94%, melting point 114.7°C, IR: ν_{max}/cm^{-1} 2072 (N=C=S), 1615 (C=N), 879 (C–Cl). ^{1}H-NMR (DMSD-d_6, D_2O) δ: 6.9, 8.9 (2d, 2H, 2CH pyridine, J=6.8 Hz), 7.2–8.4 (m, 3H, Ar-H). ^{13}C-NMR (DMSD-d_6) δ: 118.8, 126.6, 127.7, 129.8, 130.6, 136.4, 138.9, 140.4, 150.9, 152.0. *Anal.* Calcd for $C_{10}H_5ClN_2S$ (220.68): C, 54.43; H, 2.28; N, 12.69. Found: C, 54.67; H, 2.60; N, 12.44.

General Procedure for the Synthesis of Carbamimidothioic Acid Derivatives 19–25 A mixture of **18** (2.20 g, 0.01 mol) and the appropriate sulfonamide (0.012 mol) in dry DMF (30 mL) containing a catalytic amount of triethylamine was heated under reflux for 18h. The obtained solid was filtered and crystallized from ethanol to give **19**–**25**, respectively.

(*E*)-*N*′-(7-Chloroquinolin-4-yl)-*N*-(4-sulfamoylphenyl)-carbamimidothioic Acid (**19**): Yield 79%, melting point 260.4°C, IR: ν_{max}/cm^{-1} 3387, 3260, 3200 (NH, NH_2), 3095 (CH arom.), 1612 (C=N), 1329, 1155 (SO_2), 815 (C–Cl). ^{1}H-NMR (DMSD-d_6, D_2O) δ: 2.5 (s, 1H, SH, D_2O exchangeable), 6.5, 8.6 (2d, 2H, 2CH pyridine, J=6.7 Hz), 7.2–8.1 (m, 8 H, Ar-H+NH), 8.4 (s, 2H, SO_2NH_2, D_2O exchangeable) ^{13}C-NMR (DMSD-d_6) δ: 115.7, 119.0, 123.3, 126.8 (2), 126.9, 127.0, 127.2, 127.9, 132.1, 138.7, 144.7, 151.3, 152.3, 156.0, 165.3. *Anal.* Calcd for $C_{16}H_{13}ClN_4O_2S_2$ (392.88): C, 48.91; H, 3.34; N, 14.26. Found: C, 48.66; H, 3.60; N, 14.59.

(*E*)-*N*′-(7-Chloroquinolin-4-yl)-*N*-(4-(*N*-(3-methylisoxazol-5-yl)sulfamoyl)phenyl)carbamimidothioic Acid (**20**): Yield 77%, melting point 189.0°C, IR: ν_{max}/cm^{-1} 3348, 3290 (NH), 3095 (CH arom.), 2970, 2865 (CH aliph.), 1616 (C=N), 1375, 1161 (SO_2), 821 (C–Cl). ^{1}H-NMR (DMSD-d_6, D_2O) δ: 2.2 (s, 1H, SH, D_2O exchangeable), 2.3 (s, 3H, CH_3), 6.1 (s, 1H, CH isoxazole), 6.6, 8.6 (2d, 2H, 2CH pyridine, J=7.0 Hz), 7.2–8.3 (m, 8 H, Ar-H+NH), 8.7 (s, 1H, SO_2NH, D_2O exchangeable). ^{13}C-NMR (DMSD-d_6) δ: 12.0, 102.5, 115.8, 118.2 (2), 123.7, 126.6 (2), 127.5, 128.0, 128.4, 128.7, 138.1, 144.3, 146.9, 153.2, 157.9, 160.2, 162.2, 169.5. *Anal.* Calcd for $C_{20}H_{16}ClN_5O_3S_2$ (473.96): C, 50.68; H, 3.40; N, 14.78. Found: C, 50.40; H, 3.12; N, 14.98.

(*E*)-*N*′-(7-Chloroquinolin-4-yl)-*N*-(4-(*N*-(3,4-dimethylisoxazol-5-yl)sulfamoyl)phenyl)carbamimidothioic Acid (**21**): Yield 79%, melting point 182.2°C, IR: ν_{max}/cm^{-1} 3348, 3215 (NH), 3085 (CH arom.),, 2966, 2842 (CH aliph.), 1620 (C=N), 1375, 1155 (SO_2), 819 (C–Cl). ^{1}H-NMR (DMSD-d_6, D_2O) δ: 2.1 (s, 1H, SH, D_2O exchangeable), 2.2, 2.3 (2s, 6H, 2CH_3), 6.7, 8.6 (2d, 2H, 2CH pyridine, J=6.9 Hz), 7.0–8.1 (m, 8 H, Ar-H+NH), 8.9 (s, 1H, SO_2NH, D_2O exchangeable). ^{13}C-NMR (DMSD-d_6) δ: 9.2, 13.2, 102.6, 115.2, 116.7 (2), 124.0, 126.1 (2), 126.3, 126.9, 127.4, 128.1, 134.0, 148.3, 150.7, 152.3, 157.6, 158.4, 159.1, 163.9. *Anal.* Calcd for $C_{21}H_{18}ClN_5O_3S_2$ (487.98): C, 51.69; H, 3.72; N, 14.35. Found: C, 51.96; H, 3.48; N, 14.10.

(*E*)-*N*′-(7-Chloroquinolin-4-yl)-*N*-(4-(*N*-thiazol-5-ylsulfamoyl)phenyl)carbamimidothioic Acid (**22**): Yield 80%, melting point 158.5°C, IR: ν_{max}/cm^{-1} 3400, 3370 (NH), 3100 (CH arom.), 1595 (C=N), 1389, 1138 (SO_2), 823 (C–Cl). ^{1}H-NMR (DMSD-d_6, D_2O) δ: 2.3 (s, 1H, SH, D_2O exchangeable), 6.7, 8.6 (2d, 2H, 2CH pyridine, J=6.9 Hz), 7.0–8.1 (m, 10 H, Ar-H+NH), 8.8 (s, 1H, SO_2NH, D_2O exchangeable). ^{13}C-NMR (DMSD-d_6) δ: 112.4, 116.6 (2), 119.7, 124.0, 125.1 (2), 126.6, 127.0, 127.7, 127.8, 136.9, 141.1, 145.3, 147.8, 152.2, 155.0, 160.0, 168.7. *Anal.* Calcd for $C_{19}H_{14}ClN_5O_2S_3$ (475.99): C, 47.94; H, 2.96; N, 14.71. Found: C, 47.63; H, 2.77; N, 15.08.

(*E*)-*N*′-(7-Chloroquinolin-4-yl)-*N*-(4-(*N*-pyridin-2-ylsulfamoyl)phenyl)carbamimidothioic Acid (**23**): Yield 86%, melting point 150.3°C, IR: ν_{max}/cm^{-1} 3350, 3215 (NH), 3057 (CH arom.), 1629 (C=N), 1394, 1134 (SO_2), 817 (C–Cl). ^{1}H-NMR (DMSD-d_6, D_2O) δ: 2.5 (s, 1H, SH, D_2O exchangeable), 6.7, 8.5 (2d, 2H, 2CH pyridine, J=7.4 Hz), 6.8–8.1 (m, 12 H, Ar-H+NH), 8.9 (s, 1H, SO_2NH, D_2O exchangeable). ^{13}C-NMR (DMSD-d_6) δ: 112.1, 113.4, 115.6, 118.7 (2), 124.4, 127.6 (2), 127.8, 128.3, 128.5, 128.8, 138.6, 140.1, 146.8, 151.9, 152.2, 152.7, 152.9, 154.4, 160.1. *Anal.* Calcd for $C_{21}H_{16}ClN_5O_2S_2$ (469.97): C, 53.67; H, 3.43; N, 14.90. Found: C, 53.44; H, 3.10; N, 14.68.

(*E*)-*N*′-(7-Chloroquinolin-4-yl)-*N*-(4-(*N*-pyrimidin-2-ylsulfamoyl)phenyl)carbamimidothioic Acid (**24**): Yield 88%, melting point 207.9°C, IR: ν_{max}/cm^{-1} 3431, 3390 (NH), 3085 (CH arom.), 1620 (C=N), 1340, 1155 (SO_2), 815 (C–Cl). ^{1}H-NMR (DMSD-d_6 D_2O) δ: 2.3 (s, 1H, SH, D_2O exchangeable), 6.7, 8.5 (2d, 2H, 2CH pyridine, J=7.0 Hz), 6.8–8.3 (m, 11 H, Ar-H+NH), 8.7 (s, 1H, SO_2NH, D_2O exchangeable). ^{13}C-NMR (DMSD-d_6) δ: 112.1, 115.4, 118.5 (2), 124.8, 127.6 (2), 128.5, 129.3, 129.7, 137.9, 140.9, 153.0, 154.6, 157.2, 158.3 (2), 161.9, 168.3. *Anal.* Calcd for $C_{20}H_{15}ClN_6O_2S_2$ (470.96): C, 51.01; H, 3.21; N, 17.84. Found: C, 51.34; H, 3.55; N, 17.56.

(*E*)-*N*′-(7-Chloroquinolin-4-yl)-*N*-(4-(*N*-(4-methylpyrimidin-2-yl)sulfamoyl)phenyl)carbamimidothioic Acid (**25**): Yield 84%, melting point 224.9°C, IR: ν_{max}/cm^{-1} 3350, 3210 (NH), 3065 (CH arom.), 2940, 2860 (CH aliph.), 1610 (C=N), 1375, 1151 (SO_2), 829 (C–Cl). ^{1}H-NMR (DMSD-d_6, D_2O) δ: 2.2 (s, 3H, CH_3), 2.3 (s, 1H, SH, D_2O exchangeable), 6.6, 8.2 (2d, 2H, 2CH pyridine, J=6.7 Hz), 6.7–8.1 (m, 10H, Ar-H+NH), 8.6 (s, 1H, SO_2NH, D_2O exchangeable). ^{13}C-NMR (DMSD-d_6) δ: 22.8, 111.8, 113.7, 116.2 (2), 118.8, 124.8 (2), 125.5, 127.1, 127.6, 129.3, 134.1, 143.1, 145.6, 152.8, 154.9, 156.2, 156.6, 160.1, 167.2 . *Anal.* Calcd for $C_{21}H_{17}ClN_6O_2S_2$ (484.98): C, 52.01; H, 3.53; N, 17.33. Found: C, 51.73; H, 3.36; N, 17.68.

Molecular Docking All the molecular modeling studies were carried out on an Intel Pentium 1.6 GHz processor, 512 MB memory with Windows XP operating system using Molecular Operating Environment (MOE, 10.2008) software. All the minimizations were performed with MOE until a RMSD gradient of 0.05 kcal mol^{-1} Å^{-1}with MMFF94X force field and the partial charges were automatically calculated. The X-ray crystallographic structure of PI3K complexe with its ligand (PDB ID: 3S2A) was obtained from the protein data bank. The enzyme was prepared for docking studies where: (i)

Ligand molecule was removed from the enzyme active site. (ii) Hydrogen atoms were added to the structure with their standard geometry. (iii) MOE Alpha Site Finder was used for the active sites search in the enzyme structure and dummy atoms were created from the obtained alpha spheres. (iv) The obtained model was then used in predicting the ligand enzymes interactions at the active site

Biological Screening. *In Vitro* Antitumor Activity Human breast cancer cell line (MDA-MB231), skin cancer cell line (HT 1080) and neuroblastoma cell line (SH-SY5Y) were used in this study. The cytotoxic activity was measured *in vitro* for the newly synthesized compounds using the Sulfo-Rhodamine-B stain (SRB) assay using the method of Skehan *et al.*[31] The *in vitro* anticancer screening was done by the pharmacology unit at Pharmacognosy Department, College of Pharmacy, King Saud University, Riyadh, Saudi Arabia.

Cells were plated in 96-multiwell plate (10^4 cells/well) for 24 h before treatment with the compound(s) to allow attachment of cell to the wall of the plate. Test compounds were dissolved in dimethyl sulfoxide. Different concentrations of the compound under test (5, 12.5, 25, 50 μM) were added to the cell monolayer. Triplicate wells were prepared for each individual concentration. Monolayer cells were incubated with the compound(s) for 48 h at 37°C and in atmosphere of 5% CO_2. After 48 h, cells were fixed, washed and stained for 30 min with 0.4% (wt/vol) SRB dissolved in 1% acetic acid. Excess unbound dye was removed by four washes with 1% acetic acid and attached stain was recovered with Trise–ethylenediaminetetraacetic acid (EDTA) buffer. Color intensity was measured in an enzyme-linked immunosorbent assay (ELISA) reader. The relation between surviving fraction and drug concentration is plotted to get the survival curve for cancer cell line after the specified time. The molar concentration required for 50% inhibition of cell viability (IC_{50}) was calculated and compared to the reference drug Doxorubicin (CAS, 25316-40-9) and the results are given in Table 2.

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