含苯并咪唑的 2,4-取代喹唑啉衍生物的合成及抗肿瘤活性评价

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摘要 为了寻找高效的抗肿瘤药物, 设计并合成了一系列新型的 2,4-取代喹唑啉类衍生物, 采用噻唑蓝(MTT)法对目标化合物在人类胃癌细胞(MGC-803)、乳腺癌细胞(MCF-7)和人正常胃黏膜上皮细胞 GES-1 进行抗肿瘤活性评价, 结果显示部分化合物对 MGC-803 和 MCF-7 表现出中度至强效的抗肿瘤活性. 喹唑啉的 4 位被不同芳胺取代时, 2-(((1H-苯并[d]咪唑-2-基)甲基)硫基)-N-(4-甲氧基苯基)喹唑啉-4-胺(**15e**)对 MGC-803 具有较好的抗肿瘤活性, IC$_{50}$ 值为 4.60 μmol•L^{-1}; 喹唑啉的 4 位被不同查尔酮取代时, (E)-1-(4-((2-(((1H-苯并[d]咪唑-2-基)甲基)硫基)喹唑啉-4-基)氨基)苯基)-3-(3-硝基苯基)丙-2-烯-1-酮(**15k**)对 MGC-803 具有很强的抗肿瘤活性, IC$_{50}$值为 0.97 μmol•L^{-1}, 明显优于化合物 **15e**. 但是化合物 **15e** 对 GES-1 的毒性远远大于化合物 **15k**, 化合物 **15k** 的毒性与对照药品 5-氟尿嘧啶和吉非替尼相近. 分子对接结果显示, 化合物 **15k** 与表皮生长因子受体(EGFR)的结合模式优于 **15e**, 为研究新型的 EGFR 抑制剂提供了新的思路.

关键词 喹唑啉; 苯并咪唑; 合成; 抗肿瘤活性

Synthesis and Antitumor Evaluation of 2,4-Substituted Quinazoline Derivatives Containing Benzimidazole

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Abstract In order to find more efficient and economical antitumor drugs, a series of novel 2,4-substituted quinazoline derivatives containing benzimidazole were designed, synthesized and evaluated for their antitumor activities on two human tumor cell lines including human gastric cancer cells (MGC-803), human breast cancer cells (MCF-7) and the normal human gastric epithelial cell line (GES-1) by using thiazolyl blue tetrazolium bromide (MTT) assay *in vitro*. Among all the tested compounds, some compounds displayed moderate to potent antitumor activities against MGC-803 and MCF-7. When the 4-position of quinazoline was substituted by different aromatic amines, 2-(((1H-benzo[d]imidazol-2-yl)methyl)thio)-N-(4-methoxyphenyl)quinazolin-4-amine (**15e**) had good anti-tumor activity against MGC-803 with IC$_{50}$ value of 4.60 μmol•L^{-1}. When the 4-position of quinazoline was replaced by different chalcone, (E)-1-(4-((2-(((1H-benzo[d]imidazol-2-yl)methyl)thio)quinazolin-4-yl)amino)phenyl)-3-(3-nitrophenyl)prop-2-en-1-one (**15k**) had a strong anti-tumor activity against MGC-803 with IC$_{50}$ value of 0.97 μmol•L^{-1}, which was significantly better than compound **15e**. However, the toxicity of compound **15e** was more serious than compound **15k** whose toxicity was similar to that of the control drug 5-fluorouracil and gefitinib against GES-1. The result of docking with epidermal growth factor receptor (EGFR) suggested that the binding mode of **15k** was better than that of **15e**. It is believed that this work would be very useful for developing a new series of EGFR inhibitors.

1　Introduction

Nowadays, cancer is the second common cause of death, only behind the incidence of heart disease in the USA. A total of 1658370 new cancer cases and 589430 cancer deaths were projected to occur in the USA in 2015.[1,2] The World Health Organization (WHO) estimates that the incidence of cancer will reach to 22 million per year within the next two decades.[3,4] Therefore, it is imminent to research and develop effective antitumor drugs.

In the path of identifying various chemical substances which may serve as leads for designing novel antitumor agents, nitrogen-containing heterocycles are of particular interest. For example, quinazoline is considered an effective group which has good anti-malarial,[5,6] anti-cancer,[7] anti-bacterial,[8] anti-spasm, anti-inflammatory and other biological activities. Its derivatives are potent inhibitors of epidermal growth factor receptor (EGFR), such as in Figure 1. Compounds gefitinib (**1**) and lapatinib (**2**) as the EGFR inhibitors were highly effective in the treatment of lung-cancer and breast cancer, which are 4-anilinoquinazoline derivatives. In 2017 Xiao *et al.*[9] reported that 2-substituted quinazoline (**3**) had significant inhibitory effect on MDA-MB-231 cell, and its GI_{50} was 1.60 μmol·L^{-1}. In 2014 Gottasová *et al.*[10] reported the *in-vitro* activity of 4-aniloquinazoline **4** against *B. subtilis* and *S. aureus*, an EC_{50} of 0.8 μg/mL against *S. aureus* and an EC_{50} of 0.7 μg/mL in assays using *B. subtilis*. In 2017 Cui *et al.*[11] reported that compound **5** exhibited extremely high antiproliferative activity in the NIH-NCI 60 human tumor cell line panel, with low to sub-nanomolar GI_{50} values (10^{-10} mol·L^{-1} level).

In addition, benzimidazole derivatives have a very wide range of biological activity,[12,13] which were found that 2-substituted benzimidazole plays an important role in antitumor derivatives, such as compounds **6** and **8** (Figure 2). In 2010, Refaat[14] reported that many 2-substituted benzimidazole derivatives were synthesized and have significant inhibitory effects on HepG-2 and MCF-7 cells, and their IC_{50} is less than 10 μg/mL.[15,16]
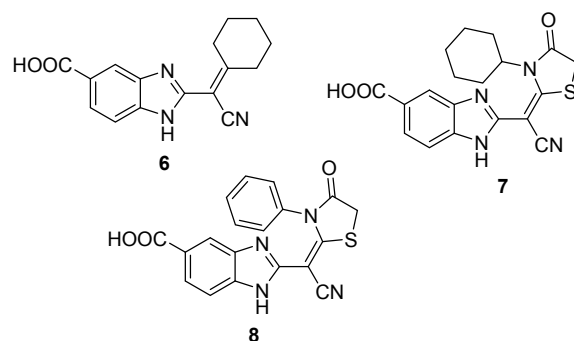
Figure 2　Chemical structures of benzimidazole compounds **6**~**8**

Based on the importance of quinazoline and benzimidazole moiety for anticancer activity, it is considered that simultaneous administration of quinazoline and benzimidazole structures in a single molecule would lead to a potent antitumor activity. So a series of novel 2,4-substituted quinazoline derivatives containing benzimidazole were designed through assignment different group (Scheme 1). The Lipinski parameters of these virtual compounds were predicted by SYBYL-X 2.0. The results showed that the most of these compounds are match the conditions of Lipinski rule. Thereby they were synthesized and evaluated on antitumor activities *in vitro*.
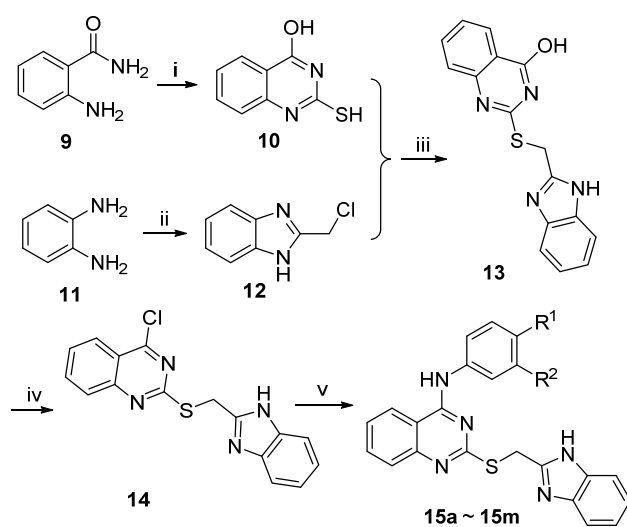
Scheme 1　Synthesis of 2,4-substituted quinazoline derivatives containing benzimidazole

Figure 1　Chemical structures of quinazoline compounds **1**~**5**

2 Results and discussion

2.1 Chemistry

The synthetic strategy to prepare the target compounds was depicted in Scheme 1. A series of novel 2,4-substituted quinazoline derivatives containing benzimidazole were finally obtained in five steps. In this letter,[17~20] we synthesized compound **10** in a higher yield under the condition of reflux in the presence of potassium hydroxide and excess carbon disulfide in ethanol at 90 ℃ for 5 h and compound **12** in a higher yield under the condition of reflux in the presence of *O*-phenylenediamine and chloroacetyl chloride in hydrochloric acid at 110 ℃ for 5 h. The next part of this synthesis involved the preparation of compound **13** which was stirred by condensation of compounds **10** and **12** in the mixed solvent of water and acetone, then compound **13** was added to phosphorus oxychloride and then slowly heated to 90 ℃ for 2 h to give compound **14**. Next, compound **14** and appropriate aniline or chalcone were added to isopropanol and the temperature was raised to 90 ℃. When the reaction was completed, filtered, the cake was washed with ethanol or by silica gel column chromatography to get the target compounds **15a**~**15m**. All the structures were fully characterized by ¹H NMR, ¹³C NMR and HRMS.

2.2 Cytotoxic activity

All synthesized compounds were evaluated for the cytotoxic activities against two human cancer cell lines, MCF-7 (human breast cancer cell line), MGC-803 (human gastric cancer cell line) and GES-1 (human gastric mucosal cells) using MTT (3,4,5-dimethyl-2-thiazolyl)-2,5-diphenyl-2-*H*-tetrazolium bromide) assay method.[21~23] 5-Fluorouracil (5-Fu) and Gefitinib were used as reference compounds. Table 1 reported the IC₅₀ (μmol•L⁻¹) values of the tested compounds **15a**~**15m** and the standard.

From the screening results in Table 1, most of the prepared compounds exhibited significant antitumor activity and weak toxicity on all the most cell lines. Compared the antitumor activity prepared compounds against MCF-7 and MGC-803, the activities of the compounds were better against MGC-803 than MCF-7. Compared **15a**~**15d** and **15f**~**15g** with **15e**, when the 4-position of quinazoline derivatives was linked to different anilines, the benzene ring was introduced electron-withdrawing group, their antitumor activities were lower than those of electron-donating group against MCF-7 and MGC-803. The toxicity of compounds **15a**~**15e** were lower than those of compounds **15f**~**15g** against GES-1. It proved that the changes of R¹ were important for reducing the toxicity of compounds. So we designed and synthesized the compounds **15h**~**15m**.

Obviously, the toxicity of compounds **15a**~**15g** were lower than those of compounds **15h**~**15m** against GES-1, especially compounds **15k**~**15m** whose toxicities were similar to 5-Fu and Gefitinib. It proved that 4-position of the quinazoline derivatives linking to different chalcones

were better for reducing toxicity than different anilines. The antitumor activities of compounds **15h**~**15m** were better than those of 5-Fu against MGC-803. At the same time, when 4-position of the quinazoline derivative was linked to different chalcones, compared **15h** and **15i**~**15m**, when the electron-withdrawing groups were introduced into the benzene ring, the antitumor activity of the compounds was worse than no-substituted against MCF-7. Compared **15j** and **15m**, when the volume of group in the benzene ring was big, the antitumor activity will be good against MCF-7 and MGC-803. The antitumor activity and toxicity of compounds **15k** and **15l** against MGC-803 were significantly better than those of 5-Fu and Gefitinib.
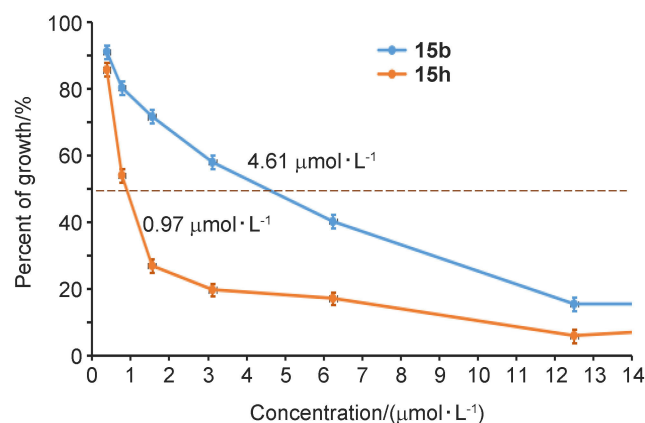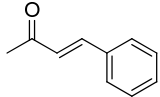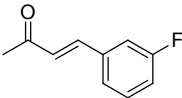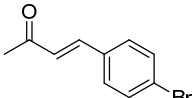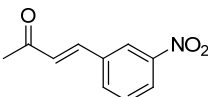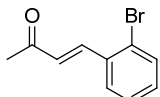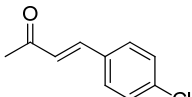

**Figure 3** Dose-dependent antiproliferative activities of **15e** and **15k** in MGC-803

The cells were incubated with increasing concentrations of compounds **15e** and **15k** for 72 h. Then the number of viable cells was quantified spectrophotometrically by the MTT reagent

In order to analyze the cause of the difference, molecular docking of **15e**, **15k** to ATP binding site of EGFR kinase was performed using Surflex-Dock module of SYBYL-X 2.0 (Figure 4). Here the EGFR complex crystal structure (3w2s.pdb) was selected as the binding model due to the similarity of core structure between **15e** and **15k**. To check the consistency, the original ligands **15e** and **15k** were docked back into the optimized receptor protein. The calculated root-mean-square deviation (RMSD) between the best docked pose and the observed X-ray crystallographic conformation of **15e** and **15k** was less than 1.50 Å. It suggested that the established docking mode is reliable. The docking results that showed compounds **15e** and **15k** were in similar binding mode as shown in Figure 4. The N(8) atom in **15e** was hydrogen-bonded to the main chain NH of the hinge region Thr854, and the length of hydrogen bond was 2.781 Å, which was a little bit longer than that of **15k** (2.705 Å). While the residue Lys745 was directly involved in hydrogen bonding with the N(14) in **15e** and **15k**, and the distance of hydrogen bond of **15e** was 1.888 Å, and a little longer than that of **15k** (1.850 Å). In addition, the chain of chalcone was inherently flexible, the residue Cys797 was directly involved in hydrogen bonding with NO₂ in **15k**. These changes should be corresponding to their different inhibitory activity on EGFR.

Table 1 Antitumor activity [$IC_{50}/(\mu mol \cdot L^{-1})$] of target compounds **15a～15m**

Compd.	R^1	R^2	MCF-7	MGC-803	GES-1
15a	F	H	53.29±1.72	24.55±1.39	>100
15b	Cl	H	52.92±1.72	15.14±1.18	>100
15c	Br	H	17.70±1.24	8.92±0.95	23.64±1.13
15d	CF_3	H	13.85±1.14	6.47±0.81	13.87±0.21
15e	OCH_3	H	2.01±1.41	4.61±0.66	8.99±0.47
15f	H	Cl	16.78±1.22	17.01±1.23	6.21±1.26
15g	H	CF_3	15.80±1.20	11.56±1.06	4.18±0.62
15h	(chalcone, phenyl)	H	15.76±1.07	4.97±0.69	38.30±2.35
15i	(chalcone, 3-F-phenyl)	H	23.24±1.00	4.65±0.66	39.37±2.17
15j	(chalcone, 4-Br-phenyl)	H	31.65±1.67	6.27±0.79	47.56±1.23
15k	(chalcone, 3-NO_2-phenyl)	H	53.88±0.12	0.97±0.19	90.55±0.31
15l	(chalcone, 2-Br-phenyl)	H	45.20±1.89	2.38±0.37	>100
15m	(chalcone, 4-Cl-phenyl)	H	61.10±1.60	7.06±1.12	>100
5-Fu[b]	—	—	9.11±0.17	7.13±0.28	77.08±1.60
Gefitinib[c]	—	—	7.34±0.87	3.98±0.33	89.35±1.32

[a] Inhibitory activity was assayed by exposure for 72 h to substances and expressed as concentration required to inhibit tumor cell proliferation by 50% (IC_{50}). [b] Positive control. [c] Positive control.
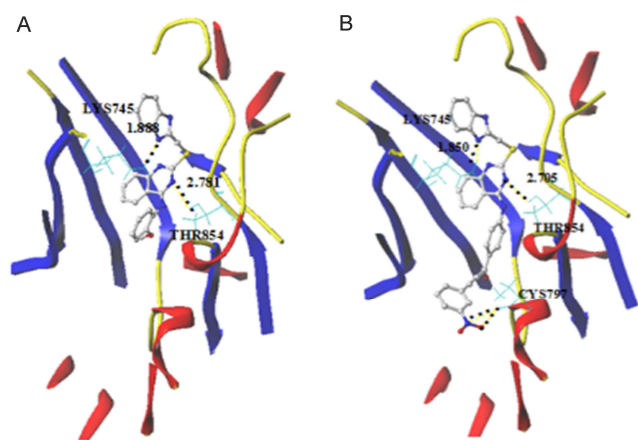
Figure 4 Binding modes of EGFR and compounds **15e** (A) or **15k** (B) docked by the Surflex-Dock program
The hydrogen bonds were illustrated as black dashed lines (length unit in Å)

3 Conclusion

In summary, a series of novel 2,4-substituted quinazoline derivatives containing benzimidazole were designed, synthesized and evaluated for their antitumor ability against two human cancer cell lines (MGC-803 and MCF-7) and the normal human gastric epithelial cell line (GES-1) using MTT assay *in vitro*. Some compounds displayed moderate to potent antitumor activity. Especially compounds **15k** and **15l** with the structure of chalcone exhibited extremely potent antitumor activity against MGC-803 with IC_{50} values less than 3 $\mu mol \cdot L^{-1}$. They are remarkably better than those of compounds **15e**, 5-fluorouracil and gefitinb. At the same time, the toxicities of compounds **15k** and **15l** were beter than those of 5-fluorouracil and gefitinb. The result of docking with EGFR suggested that the binding mode of **15k** was better than that of **15e**. It is suggested that 2,4-substituted quinazoline derivatives containing benzimidazole will be

an important class of potential cancer therapeutics.

4 Experimental

4.1 Materials

Silica gel: China Qingdao Ocean Chemical Group Corporation. Column chromatography silica gel: Shanghai May Fourth Chemical Reagent Factory. Acetyl chloride: Tianjin Miou Chemical Reagent Co., Ltd.; Glacial acetic acid: Tianjin Yongda Chemical Reagent Co., Ltd.; *N,N*-dimethylformamide: Guangdong Guanghua Technology Co., Ltd.; anhydrous ethanol: Yantai City, both Chemical Co., Ltd. The separation and purification of organic solvents used in this paper are industrial grade, after re-distillation use, other reagents are commercially available analysis. ^{1}H NMR and ^{13}C NMR spectra were measured using a DPX-DPX-400 superconducting nuclear magnetic resonance instrument, TMS as internal standard. High-resolution mass spectrometry was measured using a Waters-Micromass Q-TofMicro High Resolution Determination of tetragonal-flight time tandem mass spectrometer.

4.2 Chemistry

4.2.1 Synthesis of 2-mercapto-4-hydroxyquinazoline (10)

Potassium hydroxide (0.01 mol) was dissolved in 30 mL of absolute ethanol at room temperature. Carbon disulfide (0.01 mol) was added and stirred for 30 min. Compound **9** (0.01 mol) was added at 40 ℃, then the temperature was raised to 80 ℃ for 24 h. The mixture was cooled to room temperature and filtered. The filter cake was dissolved in 50 mL of water and adjusted pH 6 to 7 with dilute hydrochloric acid. The mixture was filtrated and the filter cake was washed with distilled water and ethanol to give compound **10**, yield 78.5%. m.p. 132.7～133 ℃; ^{1}H NMR (400 MHz, DMSO-d_6) δ: 7.93 (dd, J=7.9, 1.2 Hz, 1H), 7.73 (ddd, J=8.6, 7.3, 1.5 Hz, 1H), 7.36 (d, J=8.1 Hz, 1H), 7.39～7.29 (m, 1H); ^{13}C NMR (101 MHz, DMSO-d_6) δ: 174.71, 160.09, 140.84, 135.78, 127.15, 124.78, 116.59, 116.28. HRMS (ESI) calcd for C$_8$H$_6$N$_2$OS [M＋H]$^+$ 178.0201; found 178.0201.

4.2.2 Synthesis of 2-(chloromethyl)-1*H*-benzo[*d*]imidazole (12)

Compounds **12** was synthesized according to the literature (Lin *et al.*, 2002).

4.2.3 Synthesis of 2-(((1*H*-benzo[*d*]imidazol-2-yl)methyl)thio)quinazolin-4-ol (13)

Potassium hydroxide (8.00 mmol) was dissolved in 20 mL of distilled water, then compound **10** (5.00 mmol) was added under 55 ℃. When it was dissolved, the acetone (10 mL) solution of compound **12** (5.00 mmol) was added into the upper reaction solution and the reaction continued for 30 min. The mixture was filtered and the filter cake was washed with acetone and dried *in vacuo* to give compound **13**, white solid, yield 68.9%. m.p. 142.2～142.9 ℃; ^{1}H NMR (400 MHz, DMSO-d_6) δ: 8.05 (dd, J=7.9, 1.2 Hz,

1H), 7.78 (m, 1H), 7.62 (d, J=7.8 Hz, 1H), 7.51 (dd, J=5.9, 3.3 Hz, 2H), 7.44 (m, 1H), 7.16 (dd, J=6.0, 3.2 Hz, 2H), 4.76 (s, 2H); ^{13}C NMR (101 MHz, DMSO-d_6) δ: 168.20, 161.13, 154.07, 152.19, 147.97, 135.25, 134.78, 126.19, 126.09, 126.00, 125.25, 122.39, 122.13, 120.00, 115.80, 31.51. HR-MS (ESI) calcd for C$_{16}$H$_{12}$N$_4$OS [M＋H]$^+$ 308.0273; found 308.0273.

4.2.4 Synthesis of 2-(((1*H*-benzo[*d*]imidazol-2-yl)methyl)thio)-4-chloroquinazoline (14)

Under ice-cooling, compound **13** (5.00 mmol) was added to 10 mL of phosphorus oxychloride and then heated slowly to 90 ℃. After heated 2 h, the reaction was stopped. The solution was cooled to room temperature and ice water was added slowly. The mixture was filtered and the filter cake was washed with distilled water to give compound **14**, light gray solid, yield 88.9%. m.p. 152.1～152.7 ℃; ^{1}H NMR (400 MHz, DMSO-d_6) δ: 8.20 (d, J=8.2 Hz, 1H), 8.06 (d, J=7.1 Hz, 1H), 7.97 (d, J=8.3 Hz, 1H), 7.75 (t, J=7.6 Hz, 1H), 7.50 (dd, J=5.6, 3.2 Hz, 2H), 7.18～7.12 (m, 2H), 4.79 (s, 2H); ^{13}C NMR (101 MHz, DMSO-d_6) δ: 168.24, 161.20, 154.18, 152.14, 147.82, 135.22, 134.78, 126.20, 126.08, 126.00, 125.82, 125.26, 122.36, 122.12, 119.95, 31.50. HRMS (ESI) calcd for C$_{16}$H$_{11}$ClN$_4$S [M＋H]$^+$ 326.0393; found 326.0394.

4.2.5 Synthesis of 2,4-substituted quinazoline derivatives containing benzimidazole (15a～15m)

Compound **14** (0.50 mmol) and appropriate aniline (0.60 mmol) or chalcone (0.60 mmol) were added to 4 mL of isopropanol and the temperature was raised to 90 ℃. The reaction was quenched by TLC. When the reaction was completed, the mixture was filtrated and the filter cake was washed with ethanol or purifed by silica gel column chromatography to get the target compounds **15a～15m**.

2-(((1*H*-Benzo[*d*]imidazol-2-yl)methyl)thio)-*N,N*-bis(4-fluorophenyl)quinazolin-4-amine (**15a**): white solid, yield 90.6%. m.p. 178～179 ℃; ^{1}H NMR (400 MHz, DMSO-d_6) δ: 10.18 (s, 1H), 8.53 (d, J=8.2 Hz, 1H), 7.80 (t, J=7.7 Hz, 1H), 7.71 (dd, J=6.2, 3.2 Hz, 3H), 7.69～7.63 (m, 3H), 7.54 (d, J=8.0 Hz, 1H), 7.51～7.45 (m, 2H), 7.15 (t, J=8.9 Hz, 2H), 4.87 (s, 2H); ^{13}C NMR (101 MHz, DMSO-d_6) δ: 164.20, 158.09, 152.31, 135.03, 134.24, 132.02, 126.59, 125.99, 125.76, 125.51, 125.43, 124.08, 115.63, 115.41, 114.36, 113.61, 26.75. HR-MS (ESI) calcd for C$_{22}$H$_{17}$FN$_5$S [M＋H]$^+$ 402.1188; found 402.1189.

2-(((1*H*-Benzo[*d*]imidazol-2-yl)methyl)thio)-*N,N*-bis(4-chlorophenyl)quinazolin-4-amine (**15b**): white solid, yield 86.7%. m.p. 167.4～168 ℃; ^{1}H NMR (400 MHz, DMSO-d_6) δ: 10.21 (s, 1H), 8.54 (d, J=8.2 Hz, 1H), 7.84～7.78 (m, 1H), 7.72 (dt, J=6.6, 3.0 Hz, 5H), 7.66 (d, J=8.3 Hz, 1H), 7.56～7.52 (m, 1H), 7.52～7.47 (m, 2H), 7.40～7.34 (m, 2H), 4.89 (s, 2H); ^{13}C NMR (101 MHz, DMSO-d_6) δ: 164.06, 157.91, 152.25, 150.02, 137.76, 134.33, 131.68, 128.74, 128.54, 126.58, 126.09, 125.92, 124.82, 124.28, 124.16, 114.30, 113.64, 25.91. HRMS (ESI) calcd for C$_{22}$H$_{17}$ClN$_5$S [M＋H]$^+$ 418.0893; found 418.0893.

2-(((1*H*-Benzo[*d*]imidazol-2-yl)methyl)thio)-*N,N*-bis(4-

bromophenyl)quinazolin-4-amine (**15c**): white solid, yield 80.4%. m.p. 179～180℃; ^{1}H NMR (400 MHz, DMSO-d_6) δ: 10.24 (s, 1H), 8.58 (d, J=8.3 Hz, 1H), 7.81 (t, J=7.6 Hz, 1H), 7.73 (dd, J=6.0, 2.8 Hz, 3H), 7.71～7.66 (m, 2H), 7.54 (d, J=7.9 Hz, 1H), 7.51 (s, 2H), 7.48 (dd, J=6.2, 2.7 Hz, 3H), 4.90 (s, 2H); ^{13}C NMR (101 MHz, DMSO-d_6) δ: 164.12, 157.87, 152.31, 150.18, 138.28, 134.32, 131.91, 131.69, 126.71, 126.08, 125.84, 125.10, 124.13, 116.65, 114.36, 113.71, 26.74. HRMS (ESI) calcd for $C_{22}H_{17}BrN_5S$ [M＋H]$^+$ 462.0388; found 462.0387.

2-(((1H-Benzo[d]imidazol-2-yl)methyl)thio)-N,N-bis(4-(trifluoromethyl)phenyl)quinazolin-4-amine (**15d**): white solid, yield 70.2%. m.p. 178.7～181 ℃; ^{1}H NMR (400 MHz, DMSO-d_6) δ: 10.65 (s, 1H), 8.68 (d, J=8.2 Hz, 1H), 7.97 (d, J=8.5 Hz, 2H), 7.87 (t, J=7.3 Hz, 1H), 7.73 (dt, J=6.7, 5.5 Hz, 4H), 7.65 (d, J=8.7 Hz, 2H), 7.59 (t, J=7.5 Hz, 1H), 7.54～7.48 (m, 2H), 4.96 (s, 2H); ^{13}C NMR (101 MHz, DMSO-d_6) δ: 163.87, 158.04, 151.65, 142.05, 135.16, 131.16, 130.04, 129.49, 128.74, 126.71, 126.16, 125.97, 125.93, 125.09, 124.79, 123.37, 114.22, 113.39, 26.63. HRMS (ESI) calcd for $C_{23}H_{17}F_3N_5S$ [M＋H]$^+$ 452.1157; found 452.1158.

2-(((1H-Benzo[d]imidazol-2-yl)methyl)thio)-N,N-bis(4-methoxyphenyl)quinazolin-4-amine (**15e**): white solid, yield 81.1%. m.p. 187～188 ℃; ^{1}H NMR (400 MHz, DMSO-d_6) δ: 10.78 (s, 1H), 8.64 (d, J=8.3 Hz, 1H), 7.89 (t, J=7.6 Hz, 1H), 7.80～7.69 (m, 4H), 7.60 (t, J=7.7 Hz, 1H), 7.56～7.50 (m, 2H), 7.36 (d, J=8.7 Hz, 2H), 6.70 (d, J=8.5 Hz, 2H), 4.89 (s, 2H), 3.71 (s, 3H); ^{13}C NMR (101 MHz, DMSO-d_6) δ: 163.67, 158.07, 157.36, 150.85, 135.60, 131.27, 127.02, 126.59, 126.09, 125.84, 125.02, 114.23, 113.79, 112.76, 55.58, 26.69. HRMS (ESI) calcd for $C_{23}H_{20}N_5OS$ [M＋H]$^+$ 414.1388; found 414.1390.

2-(((1H-Benzo[d]imidazol-2-yl)methyl)thio)-N,N-bis(3-chlorophenyl)quinazolin-4-amine (**15f**): white solid, yield 84.1%. m.p. 171.9～173 ℃；^{1}H NMR (400 MHz, DMSO-d_6) δ: 10.40 (s, 1H), 8.61 (d, J=8.3 Hz, 1H), 7.93 (s, 1H), 7.84 (t, J=7.5 Hz, 1H), 7.75 (dd, J=6.2, 3.1 Hz, 2H), 7.71 (t, J=7.9 Hz, 2H), 7.57 (d, J=7.9 Hz, 1H), 7.55～7.48 (m, 3H), 7.35 (t, J=8.1 Hz, 1H), 7.16 (dd, J=7.9, 1.3 Hz, 1H), 4.94 (s, 2H); ^{13}C NMR (101 MHz, DMSO-d_6) δ: 163.81, 157.99, 151.83, 140.03, 134.84, 133.08, 131.26, 130.43, 129.37, 126.51, 126.19, 125.41, 124.70, 124.51, 124.19, 122.93, 121.84, 114.29, 113.46, 26.47. HRMS (ESI) calcd for $C_{22}H_{17}ClN_5S$ [M＋H]$^+$ 418.0893; found 418.0893.

2-(((1H-Benzo[d]imidazol-2-yl)methyl)thio)-N,N-bis(3-(trifluoromethyl)phenyl)quinazolin-4-amine (**15g**): white solid, yield 75.7%. m.p. 173.5～174 ℃; ^{1}H NMR (400 MHz, DMSO-d_6) δ: 10.51 (s, 1H), 8.62 (d, J=8.2 Hz, 1H), 8.24 (s, 1H), 8.08 (d, J=8.1 Hz, 1H), 7.84 (t, J=7.5 Hz, 1H), 7.78～7.68 (m, 4H), 7.60～7.55 (m, 2H), 7.51 (dd, J=6.1, 3.1 Hz, 2H), 7.45 (d, J=7.8 Hz, 1H), 4.93 (s, 2H). ^{13}C NMR (101 MHz, DMSO-d_6) δ: 163.84, 157.96, 152.05, 139.60, 134.70, 131.23, 130.02, 129.78, 129.47, 126.77, 126.45, 126.20, 125.86, 124.28, 123.15, 121.05, 119.40, 114.27, 113.56, 26.45. HRMS (ESI) calcd for

$C_{23}H_{17}F_3N_5S$ [M＋H]$^+$ 452.1157; found 452.1155.

(*E*)-1-(4-((2-(((1H-Benzo[d]imidazol-2-yl)methyl)thio)-quinazolin-4-yl)amino)phenyl)-3-([1,1'-biphenyl]-4-yl)-prop-2-en-1-one (**15h**): white solid, yield 80.1%. m.p. 218～219 ℃; ^{1}H NMR (400 MHz, DMSO-d_6) δ: 10.36 (s, 1H), 8.62 (d, J=7.5 Hz, 1H), 8.19 (d, J=8.8 Hz, 2H), 8.05～7.99 (m, 3H), 7.94 (dd, J=6.7, 2.8 Hz, 2H), 7.85 (t, J=7.2 Hz, 1H), 7.77 (d, J=15.6 Hz, 1H), 7.71 (dd, J=6.2, 3.0 Hz, 3H), 7.58 (t, J=7.6 Hz, 1H), 7.48 (tt, J=6.2, 3.7 Hz, 5H), 4.94 (d, J=10.3 Hz, 2H); ^{13}C NMR (101 MHz, DMSO-d_6) δ: 188.00, 164.06, 157.79, 152.25, 150.38, 143.91, 143.66, 135.28, 134.50, 133.18, 131.63, 131.02, 129.83, 129.40, 129.36, 126.83, 126.25, 125.94, 124.27, 122.49, 121.94, 114.29, 113.86, 26.76. HRMS (ESI) calcd for $C_{37}H_{28}N_5OS$ [M＋H]$^+$ 514.1702; found 514.1702.

(*E*)-1-(4-((2-(((1H-Benzo[d]imidazol-2-yl)methyl)thio)-quinazolin-4-yl)amino)phenyl)-3-(3-fluorophenyl)prop-2-en-1-one (**15i**): white solid, yield 90.5%. m.p. 178～179 ℃; ^{1}H NMR (400 MHz, DMSO-d_6) δ: 10.38 (s, 1H), 8.63 (d, J=8.3 Hz, 1H), 8.21 (d, J=8.8 Hz, 2H), 8.06 (dd, J=22.5, 12.2 Hz, 3H), 7.95～7.89 (m, 1H), 7.85 (t, J=7.4 Hz, 1H), 7.79～7.68 (m, 5H), 7.61～7.44 (m, 5H), 7.32 (td, J=8.5, 2.4 Hz, 1H), 4.99 (d, J=26.3 Hz, 2H); ^{13}C NMR (101 MHz, DMSO-d_6) δ: 187.88, 164.02, 161.80, 157.79, 152.24, 150.32, 143.79, 142.43, 137.91, 137.83, 134.52, 133.00, 131.51, 131.37, 131.29, 129.93, 126.80, 126.27, 126.00, 124.28, 123.94, 121.93, 114.28, 113.86, 26.74. HRMS (ESI) calcd for $C_{31}H_{23}FN_5OS$ [M＋H]$^+$ 532.1607; found 532.1608.

(*E*)-1-(4-((2-(((1H-Benzo[d]imidazol-2-yl)methyl)thio)-quinazolin-4-yl)amino)phenyl)-3-(4-bromophenyl)prop-2-en-1-one (**15j**): white solid, yield 83.4%. m.p. 179.5～180 ℃; ^{1}H NMR (400 MHz, DMSO-d_6) δ: 10.52 (s, 1H), 8.66 (d, J=8.3 Hz, 1H), 8.15 (d, J=8.7 Hz, 2H), 8.01 (dd, J=22.8, 12.2 Hz, 3H), 7.91 (d, J=8.4 Hz, 2H), 7.85 (dd, J=10.0, 5.6 Hz, 1H), 7.76～7.65 (m, 7H), 7.57 (t, J=7.6 Hz, 1H), 7.51～7.44 (m, 2H), 4.98 (s, 2H); ^{13}C NMR (101 MHz, DMSO-d_6) δ: 187.91, 163.96, 157.82, 152.13, 149.75, 143.50, 142.56, 134.71, 134.55, 133.20, 132.36, 131.26, 131.24, 129.82, 126.43, 126.31, 126.15, 124.36, 124.30, 123.25, 122.13, 114.23, 113.73, 26.71. HRMS (ESI) calcd for $C_{31}H_{23}BrN_5OS$ [M＋H]$^+$ 592.0809; found 592.0806.

(*E*)-1-(4-((2-(((1H-Benzo[d]imidazol-2-yl)methyl)thio)-quinazolin-4-yl)amino)phenyl)-3-(3-nitrophenyl)prop-2-en-1-one (**15k**): white solid, yield 83.2%. m.p. 189～190 ℃; ^{1}H NMR (400 MHz, DMSO-d_6) δ: 10.41 (s, 1H), 8.83 (s, 1H), 8.63 (d, J=8.3 Hz, 1H), 8.40 (d, J=7.8 Hz, 1H), 8.30 (dd, J=8.1, 1.6 Hz, 1H), 8.26～8.21 (m, 3H), 8.04 (d, J=8.8 Hz, 2H), 7.91～7.69 (m, 7H), 7.58 (t, J=7.5 Hz, 1H), 7.49 (dd, J=6.1, 3.1 Hz, 2H), 4.97 (s, 1H). ^{13}C NMR (101 MHz, DMSO-d_6) δ: 187.81, 163.96, 157.80, 152.17, 149.98, 148.91, 143.82, 141.37, 137.19, 135.55, 134.61, 132.92, 131.32, 130.83, 130.03, 126.34, 126.10, 125.23, 125.06, 124.34, 123.51, 122.00, 114.26, 113.81, 26.69. HRMS (ESI) calcd for $C_{31}H_{23}N_6O_3S$ [M＋

H]$^+$ 559.1552; found 559.1555.

(*E*)-1-(4-((2-(((1*H*-Benzo[*d*]imidazol-2-yl)methyl)thio)-quinazolin-4-yl)amino)phenyl)-3-(2-bromophenyl)prop-2-en-1-one (**15l**): white solid, yield 75.6%. m.p. 180.4~180.6 ℃; ^{1}H NMR (400 MHz, DMSO-d_6) δ: 10.46 (s, 1H), 8.64 (d, *J*＝8.4 Hz, 1H), 8.28 (d, *J*＝6.6 Hz, 1H), 8.19 (d, *J*＝8.7 Hz, 2H), 8.07~7.98 (m, 4H), 7.86 (t, *J*＝7.4 Hz, 1H), 7.77 (d, *J*＝8.0 Hz, 1H), 7.71 (dd, *J*＝6.2, 3.2 Hz, 3H), 7.62~7.52 (m, 2H), 7.52~7.44 (m, 3H), 7.45~7.39 (m, 1H), 4.96 (d, *J*＝9.3 Hz, 2H); ^{13}C NMR (101 MHz, DMSO-d_6) δ: 187.76, 163.94, 157.86, 151.94, 143.55, 141.22, 134.81, 134.53, 133.77, 133.06, 132.59, 131.23, 129.89, 129.33, 128.73, 126.47, 126.10, 125.85, 125.37, 124.53, 122.27, 114.22, 113.68, 26.71. HRMS (ESI) calcd for $C_{31}H_{23}BrN_5OS$ [M＋H]$^+$ 592.0809; found 592.0809.

(*E*)-1-(4-((2-(((1*H*-Benzo[*d*]imidazol-2-yl)methyl)thio)-quinazolin-4-yl)amino)phenyl)-3-(4-chlorophenyl)prop-2-en-1-one (**15m**): white solid, yield 89.3%. m.p. 193.6~194 ℃; ^{1}H NMR (400 MHz, DMSO-d_6) δ: 10.37 (s, 1H), 8.62 (d, *J*＝8.4 Hz, 1H), 8.19 (d, *J*＝8.7 Hz, 2H), 8.08~7.95 (m, 5H), 7.85 (t, *J*＝7.5 Hz, 1H), 7.79~7.68 (m, 4H), 7.57 (t, *J*＝9.3 Hz, 3H), 7.47 (dd, *J*＝6.1, 3.1 Hz, 2H), 4.96 (s, 2H); ^{13}C NMR (101 MHz, DMSO-d_6) δ: 187.88, 164.04, 157.79, 152.23, 150.37, 143.74, 142.41, 135.46, 134.50, 134.27, 133.06, 131.60, 131.06, 129.87, 129.42, 126.83, 126.25, 125.96, 124.28, 123.25, 121.93, 114.29, 113.86, 26.75. HRMS (ESI) calcd for $C_{31}H_{23}ClN_5OS$ [M＋H]$^+$ 548.1312; found 548.1312.

Supporting Information ^{1}H NMR and ^{13}C NMR spectra of compounds **9**~**14** and **15a**~**15m** are available for free download from our website (http://sioc-jour nal.cn/).

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