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PII: S0960-894X(18)31002-3
DOI: https://doi.org/10.1016/j.bmcl.2018.12.056
Reference: BMCL 26225

To appear in: *Bioorganic & Medicinal Chemistry Letters*

Received Date: 9 October 2018
Revised Date: 17 December 2018
Accepted Date: 24 December 2018

Please cite this article as: Das, D., Xie, L., Wang, J., Xu, X., Zhang, Z., Shi, J., Le, X., Hong, J., Discovery of new quinazoline derivatives as irreversible dual EGFR/HER2 inhibitors and their anticancer activities- Part 1, *Bioorganic & Medicinal Chemistry Letters* (2018), doi: https://doi.org/10.1016/j.bmcl.2018.12.056

Discovery of new quinazoline derivatives as irreversible dual EGFR/HER2 inhibitors and their anticancer activities- Part 1

Debasis Das*, Lingzhi Xie, Jingbing Wang, Xin Xu, Zonghua Zhang, Jingli Shi, Xiaoyong Le, Jian Hong*

Discovery Chemistry Research, Arromax Pharmatech Co. Ltd.
Sangtian Island Innovation Park, No. 1 Huayun Road, Suzhou 215123, P. R. China.

*Corresponding authors:
Email: debasis.das@arromax.com (D. Das) & eric.hong@arromax.com (J. Hong).

Abstract

Overexpression of EGFR and HER2 are observed in many breast, ovarian, colon and prostate cancers. The second and third generation irreversible EGFR/HER2 dual kinase inhibitors became popular after the approval of Afatinib by FDA to overcome the mutation related problem. To find efficacious drug candidates, a series of novel quinazoline derivatives were designed, synthesized and evaluated as dual EGFR/HER2 tyrosine kinase (TK) inhibitors. Selected twenty four compounds were reported here with significant inhibitory activities against EGFR/HER2 tyrosine kinases. Several compounds showed nanomolar IC_{50} values. *In vitro* studies of quinazoline derivatives were done on NCI-H1975, HCC827, A431, MDA MB-453 cell lines. The compounds **1a**, **1d** and **1v** were found more potent compared to standard drug afatinib. *In vivo* efficacy study of **1d** on nude mice NCI-H1975 tumour xenograft model was discussed.

Keywords: EGFR, HER2, dual inhibitor, quinazoline, antitumor, anticancer, irreversible, tyrosine kinase.

The human epidermal growth factor receptor (HER) family consists of four members of tyrosine kinase (TK) receptors: HER1 (EGFR/ErbB1), HER2 (ErbB2/neu), HER3 (ErB3) and HER4 (Erb4). A complex signalling cascade occurs through homo- and hetero dimeric HER complexes formation. The downstream signalling modules after HER complex formations follow the RAS/RAF/MEK/ERK1/2, phosphatidylinositol 3-kinase and Akt (Protein kinase B) PI3K-Akt/mTOR pathways.[1,2] The downstream signalling pathways lead to cell proliferation, adhesion, survival, differentiation and apoptosis. Several malignancies, including lung, breast, stomach, colorectal, pancreatic, head & neck cancers and glioblastoma are associated with

overexpression or mutation of *HER* oncogene. Overexpression or amplification of *HER2* oncogene is observed in 25–30% of breast cancer and ovarian cancer patients.[3-5] *HER2* gene overexpression is detected in non-small cell lung cancer (NSCLC)[6], prostate cancer and prognostic factor for gastric cancer.[7, 8]

The U.S. Food and Drug Administration (FDA) approved several EGFR and HER2 targeted therapeutics in recent years. The selective EGFR tyrosine kinase inhibitors – gefitinib (Iressa, ZD1839)[9, 10] and erlotinib (Tarceva, OSI-774)[11] were approved for non-small-cell lung cancer (NSCLC) treatment. Lapatinib, a reversible dual EGFR/HER2 TK inhibitor used for breast cancer, especially for the patient who showed resistance to trastuzumab based therapy.[12,13] However, the EGFR tyrosine kinase (TK) inhibitors are limited by the up-regulation of bypass signalling pathways and acquired point mutations.[14, 15] After treatment 10–16 months with these inhibitors, approximately 50% of the NSCLC patients gained an additional gate keeper mutation (T790M) that decreases sensitivity to gefitinib or erlotinib and increases the binding affinity for ATP. After the advancement of the first generation drugs, many scientists conducted clinical studies to overcome various mutations, including EGFR and KRAS mutations.[16-18] A number of new-generation irreversible inhibitors were reported to overcome EGFR mutations (L858R and T790M) related drug resistances.[19, 20]

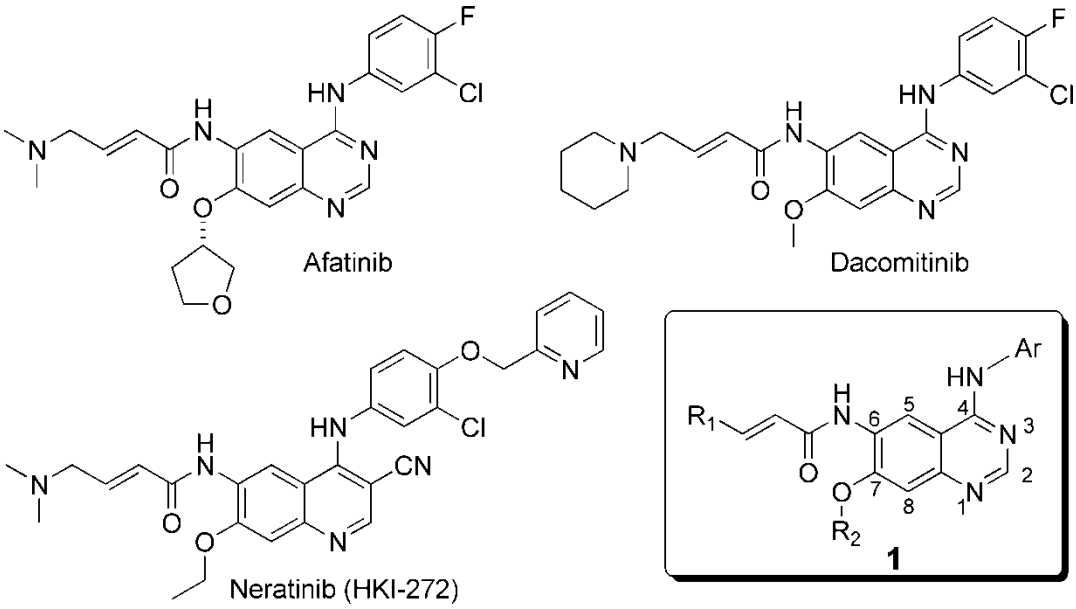
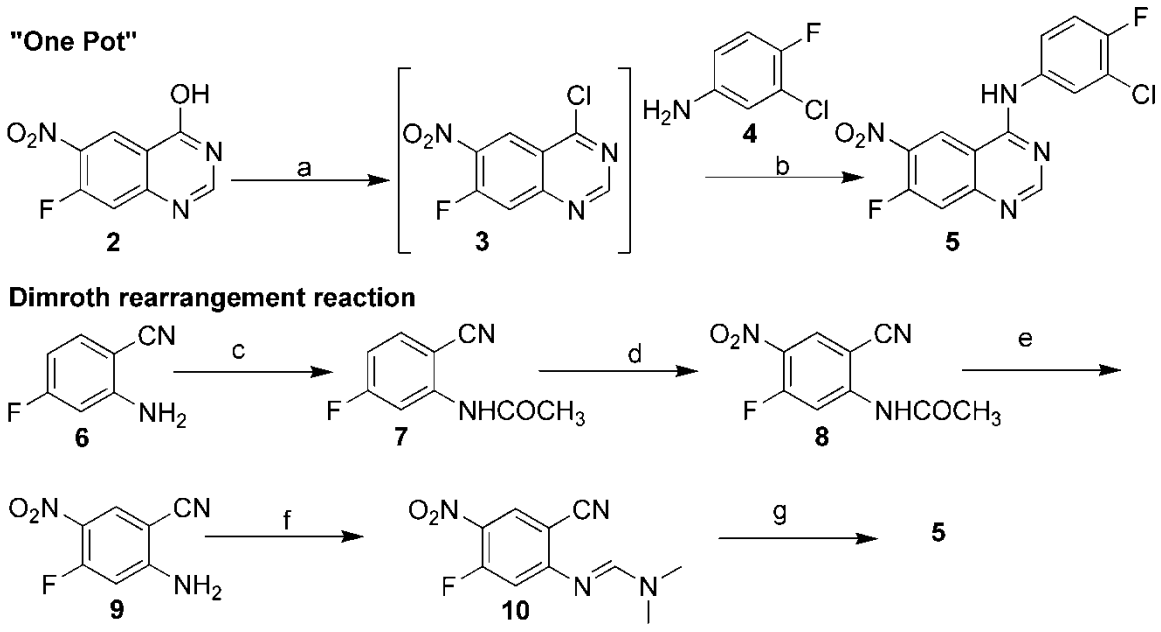


Fig. 1. Second generation irreversible EGFR/HER2-TK inhibitors.

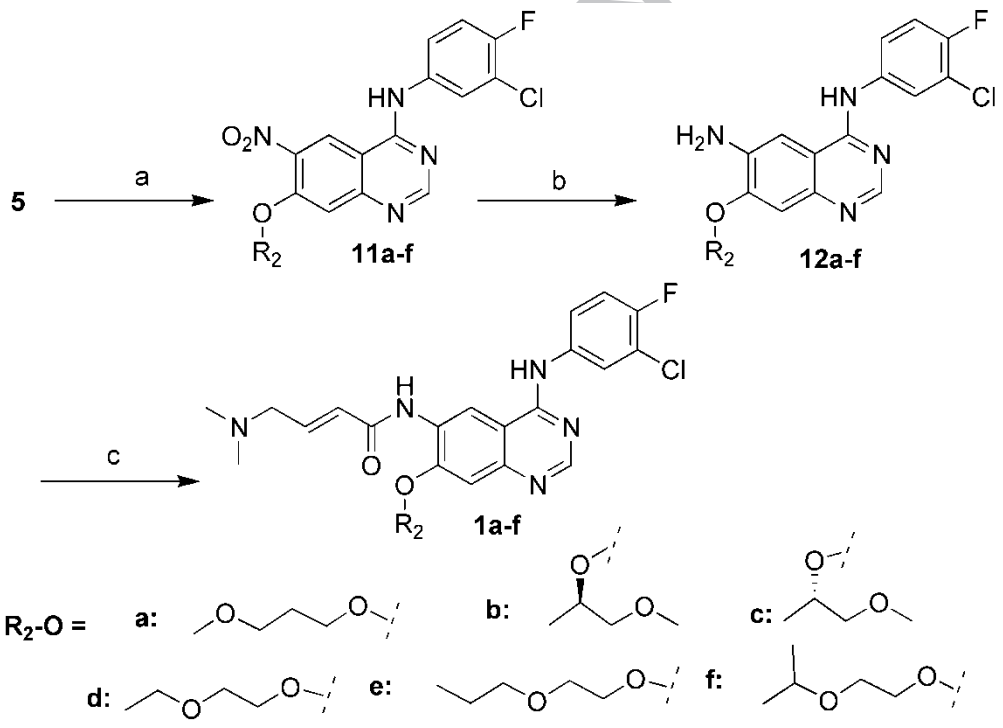
Afatinib (Gilotrif, BIBW-2992) (Figure 1), the second generation irreversible dual EGFR/HER2 inhibitor got approval by FDA for the treatment of late-stage NSCLC patients with actively mutated EGFR.[21] Erlotinib and gefitinib worked on EGFR and not worked on EGFR and KRAS mutations. Afatinib was reported to be superior to erlotinib and gefitinib on EGFR mutation. Being an EGFR/HER2 inhibitor, afatinib showed good results for the treatment of lung cancer carrying both EGFR and KRAS mutations.[22] Neratinib (HKI-272) was used for advanced HER2 positive breast cancer.[23] Dacomitinib was launched for the treatment of NSCLC.[24, 25] Identifying efficacious irreversible EGFR/HER2 dual inhibitors remain the focus of research interest for future drugs.[26]

In this article, we report the synthesis and biological evaluation of new quinazoline derivatives of general structure **1** (Figure 1). To make irreversible EGFR/HER2 dual inhibitors, a Michael acceptor was kept at the 6-position of quinazoline ring for Michael addition with the thiol group (–SH) of cysteine (Cys773) on the active center pocket wall of EGFR tyrosine kinase. The SAR studies were done by changing the size of alkoxyalkane chains at 7-position (**1a-f**), different substitutions on aryl units at 4-position (**1g-k**) and different Michael acceptors at 6-position of the quinazoline core (**1l-x**). *In vitro* pharmacological profile and *in vivo* antitumor activities of selected compounds were reported.



Scheme 1. Reagents and conditions: (a) $POCl_3$, reflux; (b) **4**, CH_3CN, reflux, overnigt, 56%; (c) Ac_2O, Et_3N, DCM, RT, 6h, 96%; (d)HNO_3, H_2SO_4, 0-20°C, 60%; (e) 37% HCl, 60°C, 2h, 70%; (f) DMF-DMA, toluene, reflux, 3h; g) **4,** 125°C, 2h, 62%.

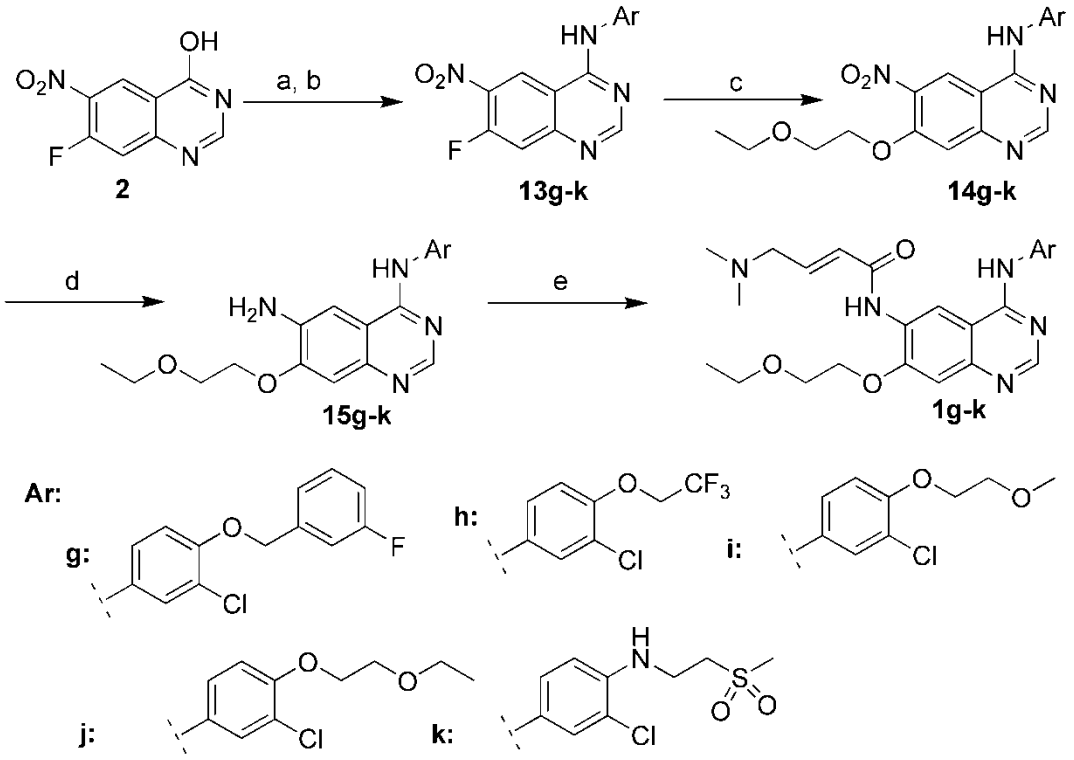
Many synthetic methods are known to construct the quinazoline derivatives.[27] The new quinazoline derivatives were made in two ways –"one pot" synthesis, starting with the qunazoline core and step-wise synthesis via Dimroth rearrangement reaction. In "One pot" synthesis, 7-fluoro-6-nitro-quinazolin-4-ol (**2**) was converted to 4-chloro-7-fluoro-6-nitro-quinazoline (**3**) using phosphorous oxychloride. Intermediate **3** was allowed to react with 3-chloro-4-fluoro-aniline (**4**) to afford quinazoline **5** (Scheme 1). In Dimroth rearrangement condition, the synthesis was started from commercially available 2-amino-4-fluoro-benzonitrile (**6**). Acetylation of **6** followed by nitration gave **8** in good yield. The hydrolysis of amidegroup of **8** was done under acidic condition and the aniline **9** was reacted with *N,N*-dimethylformamide dimethyl acetal (DMF-DMA) to give **10**. Then 3-chloro-4-fluoro-aniline (**4**) was reacted with **10** to give quinazoline **5** in moderate yield (62%).



Scheme 2. Reagents and conditions: (a) Alkoxy alkanol, NaH, DMF:THF (4:1), -15°C to rt, 1h; 58-78%; (b) $SnCl_2$, HCl or Fe/ acetic acid, (c) (E)-4-(dimethylamino)but-2-enoic acid, oxalyl chloride or thionyl chloride, DMF(cat.),THF, NMP, 1hr, then 10% NaOH to pH 11.

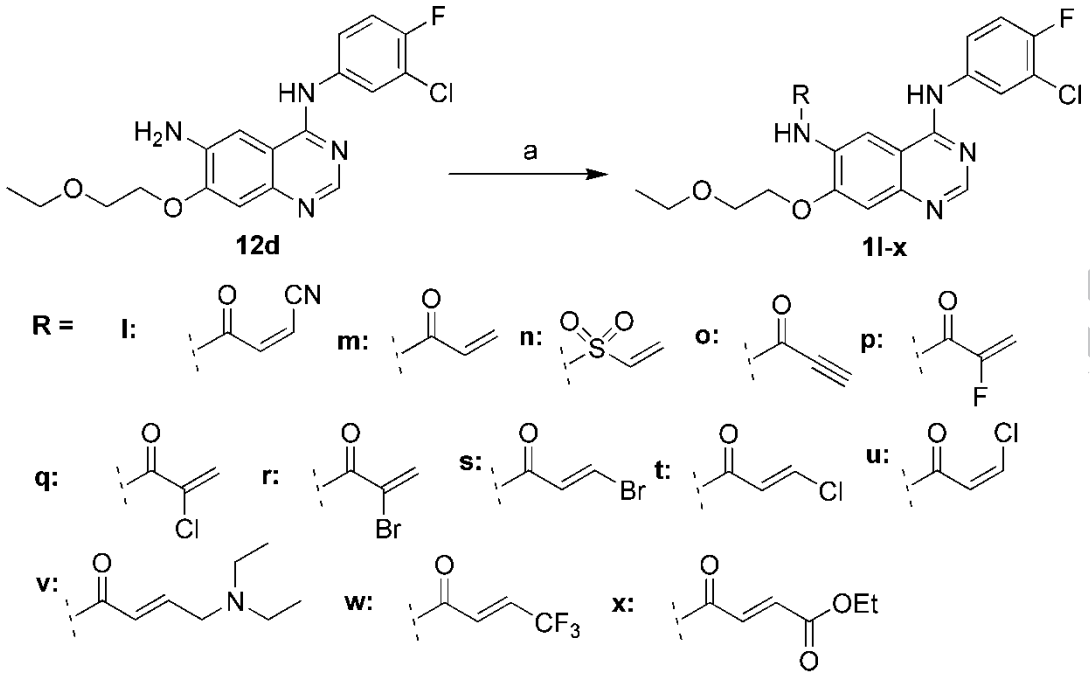
The quinazoline derivatives **1a-f** with different 7-alkoxyalkoxide chains were made in Scheme 2. In presence of sodium hydride (NaH), alkoxy alkanols (ethylene / propelene glycol methyl ether) were reacted with **5** at low temperature (–15°C to –20°C) to give 7-alkoxy quinazolines **11a-f**. The 6-nitro group of **11a-f** was easily reduced to give 7-alkoxy-6-amino

quinazolines **12a-f** by tin(II) chloride dehydrate and hydrochloric acid or iron powder in acetic acid conditions at 40°C to 60°C temperature for 30min to 3hrs. (*E*)-4-(dimethylamino)-but-2-enoyl chloride was prepared *in situ* from corresponding acid and oxalyl chloride reaction. Then (*E*)-4-(dimethylamino)but-2-enoyl chloride was reacted with **12a-f** separately to give quinazoline derivatives **1a-f** in moderate yields (46-84%). The quinazoline derivatives **1g-k** with substituted phenyls at 4-position were prepared in Scheme 3.



Scheme 3. Reagents and conditions: (a) $POCl_3$, reflux; (b) Ar-NH_2, CH_3CN, reflux, overnigt, 55-65%. (c) NaH, 2-ethoxyethanol, DMF:THF (4:1), -15°C to rt, 1h; 58-78%; (d) $SnCl_2$, HCl or Fe/ acetic acid; (e) (E)-4-(dimethylamino)but-2-enoyl chloride hydrochloride , NMP, 1h, then 10% NaOH to pH 11.

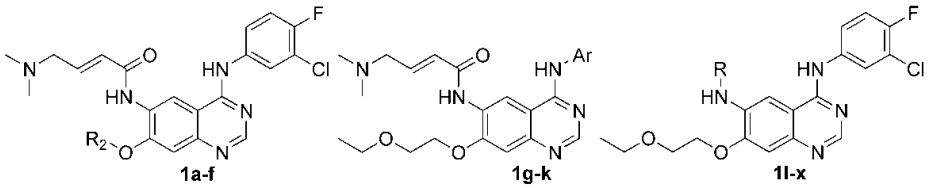
The nitroquinazolines **13g-k** were prepared with different aryl units from **2** and allowed to reacted with 2-ethoxyethanol in presence of sodium hydride to give 7-(2-ethoxyethoxy)quinazolines **14g-k** in good yields. Reduction of nitro to amines **15g-k**, followed by (*E*)-4-(dimethylamino)but-2-enoyl chloride reaction led to compounds **1g-k** in moderate yields (42-65%). The 6-aminoquninazoline **12d** was allowed to react with substituted acryloyl chlorides or vinyl sulfonyl chlorides to give compounds **1l-x** (Scheme 4).



Scheme 4. Reagents and conditions: a) Substituted crotonic acid chloride hydrochloride, NMP or Vinyl sulfonyl cholride, py.

The anti-proliferation activities of these newly synthesized twenty four compounds, **1a-x** were evaluated by cell proliferation assay kits (CCK8) (Supplementary material). *In vitro* cell proliferation were studied on three lung cancer cell lines human cell lines- human non-small cell lung cancer cell lines (NCI H1975), human non-small cell lung cancer cell line (HCC 827), human epithelial carcinoma cell lines (A431) which were harbouring EGFR T790M, L858R mutants, wild type EGFR (EGFRWT) respectively and human breast cancer cell lines (MDA-MB-453). To identify efficacious irreversible EGFR/HER2 inhibitors with more potent antitumor activities, all newly synthesized compounds were tested using afatinib as the reference standard. The data of twenty four compounds are summarized in Table 1.

Table 1. Inhibitory activities of compounds **1a-x** on different cell lines



Cell lines / Compounds		NCI-H1975	HCC 827	A431	MDA-MB-453
	CLogP	IC_{50}(nM)	IC_{50}(nM)	IC_{50}(nM)	IC_{50}(nM)
1a	5.01	36.09	0.29	23.34	N.D
1b	4.94	49.92	0.27	43.03	N.D
1c	4.94	66.50	0.34	16.70	N.D
1d	5.02	12.20	0.31	1.52	0.62
1e	5.55	10.63	0.55	8.15	N.D
1f	5.33	41.73	0.55	5.86	N.D
1g	6.51	49.66	2.54	79.49	N.D
1h	5.39	106.80	2.90	64.13	2.55
1i	4.49	7.30	3.43	20.06	1.65
1j	4.88	6.17	0.34	13.68	2.52
1k	3.38	65.60	0.53	6.48	12.70
1l	4.49	18.37	5.56	94.82	0.68
1m	4.96	51.81	0.15	6.01	5.67
1n	5.24	13.72	3.95	3.37	130.50
1o	4.09	49.31	1.46	11.45	3.42
1p	5.51	72.49	0.53	54.45	58.20
1q	6.11	37.11	33.22	368.90	3.71
1r	6.20	39.44	6.39	572.20	15.05
1s	5.30	8.21	4.86	296.60	3.99
1t	5.21	7.43	7.81	289.10	14.87
1u	5.21	1.60	7.78	24.02	N.D
1v	6.08	190.40	1.31	5.45	N.D
1w	5.25	27.56	0.15	148.20	18.30
1x	5.06	35.23	0.987	207.40	21.57
Afatinib	4.49	96.07	0.4376	10.21	N.D

N.D = Not detected, CLogP calculated on Chemdraw

By changing the alkoxy group at 7-position of the quinazoline ring, we got more potent compounds which displayed higher antiproliferative activity on H1975 cells (IC_{50} = 6.1-13.7 nM). Compound **1a** was better than afatinib in H1975 and HCC827. The chirality on the alkoxy chain (**1b & 1c**) had no significant effect on antiproliferative inhibitory effect on H1975 and HCC827. Whereas **1c** was superior almost by three fold active than **1b** for A431. Compound **1d** was better in all four cell lines. Aryl group SAR at 4-position of the quinazoline ring was done for compounds **1g-l**. It was observed that **1h** & **1l** was better for H1975 and A431, **1j** was better in H1975 compared to afatinib. Compounds containing different Michael acceptors at the 6-amino position of the quinazolines (**1m**, **n**, **o**, **v**) showed better results in H1975 and A431 cell lines.

Table 2.
In vitro inhibitory activities of the compounds against EGFR and HER2 enzymes

Compounds	IC_{50} against EGFR (nM)	IC_{50} against HER2 (nM)
1a	0.76	39.26
1b	0.94	148.30
1c	2.73	482.10
1d	0.69	42.11
1e	2.84	131.60
1f	1.54	83.84
1g	153.50	261.80
1v	1.48	10.94
Afatinib	0.96	73.72

In vitro kinase inhibitory activities for EGFR and HER2 enzymes were done for most of the synthesized quinazolines. *In vitro* kinase analysis was performed using the HTRF kinEASE TK kit of Invitrogen Co. The result of the compounds **1a-h** and **1v** were summarized in Table 2. Compounds **1a, 1d** effectively inhibited the *in vitro* kinase and showed better

activities of EGFR (IC_{50} = 0.76, 0.69 nM) and HER2 (IC_{50} = 39.2, 42.1 nM) respectively compared to afatinib (EGFR IC_{50} = 0.96 & Her2 IC_{50} = 73.72nM). The chiral variation of the alkoxy chain **1b** and **1c** were poor inhibitors for HER2. So, we kept the ethyleneglycol ethyl ether chain at C-7 position of the quinazoline scaffold and changed the substituent on the N-aryl unit. Compound **1v** was weaker in EGFR activity (IC_{50} = 1.4nM) and better for Her2 activity (IC_{50} = 10.9nM) than afatinib. In enzymatic assay, the length of the side chain played a major role in EGFR activity. The optimum chain length was observed six-atoms to show nanomolar activity, e.g. **1a**, **1d**. The longer chain lengths e.g. **1e** seven atoms and branched chain **1f** reduced activity 2.84 nM and 1.54 nM respectively. Chiral examples **1b** & **1c** showed little poorer activity than six atom lengths. **1b** was superior to **1c** by three times due to match and mismatch topology. So, six atoms linker was the optimum chain length at 7-position of quinoline for better inhibitory activity. HER2 activities of the examples were in the similar trend as EGFR. The inhibitory activity of compounds **1a** and **1d** on HER2 were better than afatinib.

Table 3. *In vivo* therapeutic effect of **1d** on NCI-H1975 nude mice xenografts

Group	**Animal No. (pcs)**	**Administration mode**	**Dosage (mg/kg)**	**RTV**	**Inhibition rate (%)**	**T/C(%)**
					d22	
Blank control	8	qd	0	7.45±1.26	---	---
Positive control (afatinib)	8	qd	20	1.67±0.20	67.6**	22.44
1d	8	qod	80	0.43±0.05	95.1***	5.75
1d	8	qod	20	1.39±0.40	82.0**	18.71
1d	8	qd	5	2.00±0.29	73.2**	26.91

Notes: (1) compared with the positive control group, *p<0.05，**p<0.01，***p<0.001. ; (2) qd: administrated every day；qod：administrated every other day；(3) d22：the time of observed 7 days after the last administration

Many compounds in the series showed good *in vitro* activities in different cell lines. However, only few of them such as compound 1d displayed good pharmacokinetics properties and bioavailability, and thus 1d was selected for further *in vivo* studies. The intravenous dosing (5mg/Kg) and PO dosing (10mg/Kg) of **1d** and its maleate salt were done on Sprague Dawley rats. The oral bioavailability of **1d** on was 46% and its malete salt was 70%. The compound **1d** showed $T_{1/2}$ = 6.8h, Tmax = 4h and Cmax = 92.3μg/L. So, the compound **1d** was selected for *in vivo* anti-tumor efficacy study.

We were interested to find active compound from this series for the mutant specific lung cancer. The human non-small cell lung cancer cell lines NCI-H1975 mice xenograft was suitable for T790M mutant studies.[28] A tumor xenograft model was established by injecting NCI-H1975 cells into nude mice of age 6-7 weeks. When tumors growth reached to 100-200 mm^3, the mices were administrated orally with **1d** at lower dose 5mg/kg daily or every other day at middle and higher doses of, 20 and 80 mg/kg, respectively. It was continuously administrated for 14 days (d14) and observed for 7 days (d22) in Table 3. Established afatinib (20 mg/kg) treated mice were included as positive controls. The negative control group was administrated with the same amount of solvent (saline solution of 1% DMSO), during the period of administration and recovery. Very good dose-dependent tumor growth inhibitions were observed with treatment of compound **1d (**Table 3). The tumor inhibition rates were observed 95.1%, 82.0%, 73.2% for high, middle and lower doses respectively. The observed tumor inhibition rates of **1d** were better than afatinib (67.6%). Middle dose of compound **1d** every other day (20mg/kg, PO, qod) was efficacious than afatinib middle dose everyday (20mg/kg, PO, qd). *In vivo* efficacy showed that the compound **1d** had better tumor inhibition effect in NCI-H1975 constructed tumors model than afatinib (Figure 2).

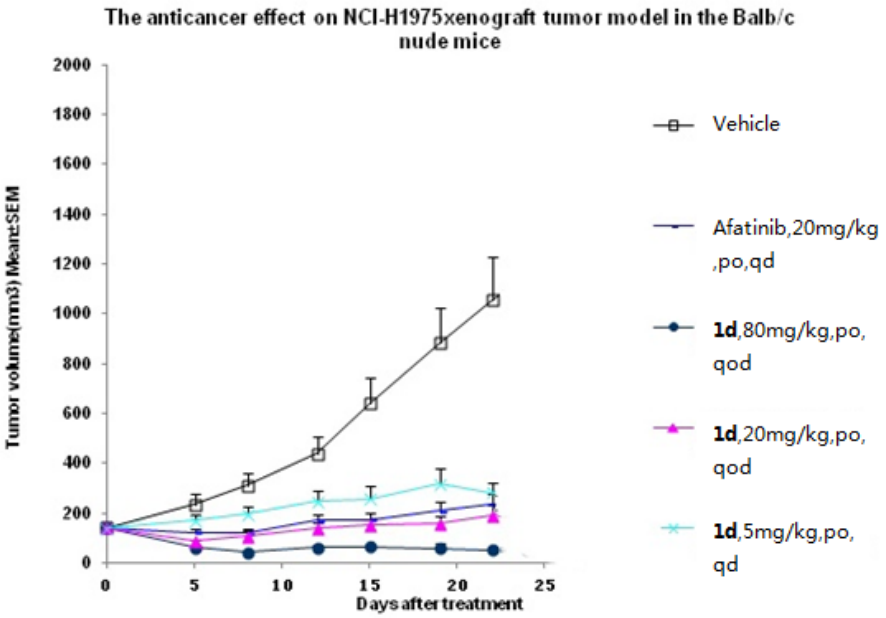


Fig. 2. *In vivo* efficacy study of **1d** in mice model.

In conclusion, a series of new quinazoline derivatives of structure **1a-x** were designed, synthesized and evaluated for bioactivity *in vitro* and *in vivo*. The synthesized compounds were tested against four human tumor cell lines (NCI-H1975, HCC 827, A431, and MDA-MB-453). A few compounds exhibited single-digit nanomolar anti-proliferation activities on all tested tumor cell lines, especially NCI-H1975 and HCC827, A431. Some of the tested derivatives displayed very good inhibition of EGFR and HER2 enzyme inhibition. Compounds **1a**, **1d** and **1v** were identified with dual EGFR/HER2 better activities than afatinib. The studies of these compounds on animal models were done. *In vivo* efficacy of **1d** was demonstrated on NCI-H1975 xenografts model. Compound **1d** showed better results compared to standard drug afatinib. Further investigations to identify the therapeutic candidate in this quinazoline series are in progress and the results will be published elsewhere.

Acknowledgments

The authors would like to acknowledge financial support from Gusu Scientific Leadership Program and Jiangsu Province Social development program, P. R. China. The authors are thankful to Shanghai Medicilon Inc. for providing full biological assay services.

Supplementary data

Synthesis, analytical data and *in vitro* and *in vivo* bioassay methods are available.

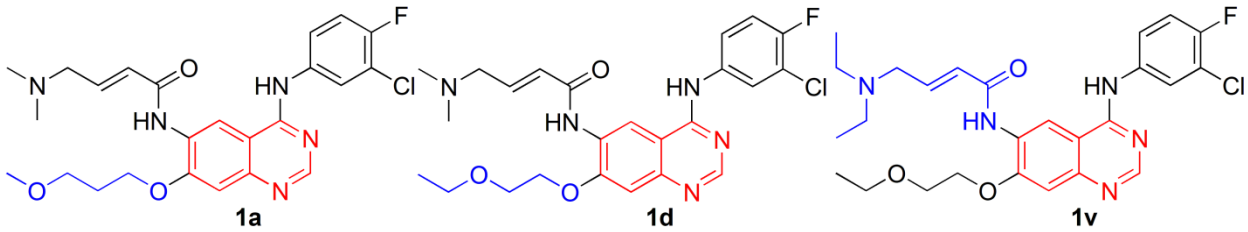
References:

1. Jordan J D, Landau E M, Iyengar R. Signaling of networks: the origin of the multicellular multi-tasking. *Cell* 2000; 103: 193-200.
2. Yarden Y, Sliwkowski M X. Untangling the ErbB signalling network. *Nat. Rev. Mol. Cell. Biol.* 2001; 2: 127-137.
3. Jackisch C, Lammers P, Jacobs, I. Evolving landscape of human epidermal growth factor receptor 2-positive breast cancer treatment and the future of biosimilars. *The Breast* 2017; 32; 199-216.
4. Ross J S, Slodkowska E A, Symmans W F et al. The HER-2 receptor and breast cancer: ten years of targeted anti-HER2 therapy and personalized medicine. *Oncologist* 2009; 14: 320–68.
5. Slamon D J, Godolphin W, Jones L A, et al. . Studies of the HER-2/*neu* proto-oncogene in human breast and ovarian cancer. *Science* (Wash. DC) 1989; 244: 707–712.
6. Cox G, Vyberg M, Melgaard B, Askaa J, Oster A, O'Byrne K J. Herceptest: HER2 expression and gene amplification in non-small cell lung cancer. *Int. J. Cancer* 2001; 92: 480–483.
7. Osman I, Scher H. I, Drobnjak M, Verbel D, Morris M, et al. HER-2/*neu* (p185) protein expression in the natural or treated history of prostate cancer. *Clin. Cancer. Res.*, 2001; 7: 2643–2647.
8. Gravalos C, Jimeno A. HER2 in gastric cancer: a new prognostic factor and a novel therapeutic target. *Ann Oncol.* 2008; 19: 1523–9.
9. Barker J, Gibson K. H, Grundy W, Godfrey A. A, et al. Studies leading to theidentification of ZD1839 (Iressa™): an orally active, selective epidermal growth factor receptor tyrosine kinase inhibitor targeted to the treatment of cancer. *Bioorg. Med. Chem. Lett.*, 2001; 11: 1911-1914.
10. Wakeling E, Guy S P, Woodburn J R, Ashton S E, Curry B J, Barker A J, Gibson K H. ZD1839 (Iressa) an orally active inhibitor of epidermal growth factor signaling with potential for cancer therapy. *Cancer Res.* 2002; 62: 5749-5754.
11. Ganjoo K N, Wakelee H. Review of erlotinib in the treatment of advanced non-small cell lung cancer. *Biol. Targets Ther. 1* 2007; 335-346
12. Burris H A. Dual kinase inhibition in the treatment of breast cancer: initial experience with the EGFR/ErbB-2 inhibitor lapatinib. *Oncologist.* 2004; 9: Suppl 3: 10–5.

13. Oxnard G R, Arcila M E, Chmielecki J, Ladanyi M, Miller V A, Pao W. New strategies in overcoming acquired resistance to epidermal growth factor receptor tyrosine kinase inhibitors in lung cancer. *Clin. Cancer Res.* 2011; 17: 5530-5537.
14. Pallis A, Briasoulis E, Linardou H, Papadimitriou C, Bafaloukos D, Kosmidis P, Murray S. Mechanisms of Resistance to Epidermal Growth Factor Receptor Tyrosine Kinase Inhibitors in Patients with Advanced Non-Small-Cell Lung Cancer: Clinical and Molecular Considerations. *Curr. Med. Chem.* 2011; 18: 1613-1628.
15. Ohashi K, Maruvka Y. E, Michor F, Pao W. Epidermal growth factor receptor tyrosine kinase inhibitor-resistant disease. *J. Clin. Oncol.* 2013; 31: 1070 − 1080.
16. Linardou H, Dahabreh I J, Kanaloupiti D. et al. Assessment of somatic k-RAS mutations as a mechanism associated with resistance to EGFR-targeted agents: a systematic review and meta-analysis of studies in advanced non-small-cell lung cancer and metastatic colorectal cancer. *Lancet Oncology*, 2008; 9: 962-972.
17. Rulli E, Marabese M, Torri V. et al. Value of KRAS as prognostic or predictive marker in NSCLC: results from the TAILOR trial. *Annals of Oncology*, 2015; 26: 2079-2084.
18. Takeda M, Okamotob I, Nakagawaa K. Pooled safety analysis of EGFR-TKI treatment for EGFR mutation-positive non-small cell lung cancer, *Lung Cancer* 2015; 88: 74-79.
19. Walter O, Tjin Tham Sjin R, Haringsma H J, Ohashi K, Sun J. et al. Discovery of a Mutant-Selective Covalent Inhibitor of EGFR that Overcomes T790M-Mediated Resistance in NSCLC. *Cancer Discov.* 2013; 3: 1404-1415.
20. Cross D A, Ashton S E, Ghiorghiu S, Eberlein C, Nebhan C A. et al AZD9291, an irreversible EGFR TKI overcomes T790M mediated resistance to EGFR inhibitors in lung cancer. *Cancer Discov.*, 2014; 4: 1046-1061.
21. Dungo, R. T, Keating, G. M. Afatinib: First Global Approval. *Drugs* 2013; 73: 1503-1515.
22. Moll H P, Pranz K, Musteanu M, Grabner B, Hruschka N, Mohrherr J, Aigner P. Afatinib restrains K-RAS–driven lung tumorigenesis. Science Translational Medicine. *Science Translational Medicine*, 2018; 10:eaao2301 DOI:10.1126/scitraslmed.aao2301).
23. Minami Y, Shimamura T, Shah K., et al. The major lung cancer-derived mutants of ERBB2 are oncogenic and are associated with sensitivity to the irreversible EGFR/ERBB2 inhibitor HKI-272. *Oncogene*. 2007; 26 (34): 5023–7.

24. Reckamp K. L, Giaccone G, Camidge D R, Gadgeel S M, Khuri F R, Engelman J A. et.al. A phase 2 trial of dacomitinib (PF-00299804), an oral, irreversible panHER (humanepidermal growth factor receptor) inhibitor, in patients with advanced non small cell lung cancer after failure of prior chemotherapy and erlotinib. *Cancer* 2014; 120: 1145-54.
25. Collisson E A, Campbell J D, Brooks A. N. et al. Comprehensive molecular profiling of lung adenocarcinoma: The cancer genome atlas research network. *Nature*, 2014; 511: 543-550.
26. Hong J, Xu X, Le X, Zhang Z. Quinazoliine derivative preparation method therefor and pharmaceutical composition and application thereof, Patent *WO2016/23217*
27. Khan I, Zaib S, Batool S, et al. Quinazolines and quinazolinones as ubiquitous structural fragments in medicinal chemistry: An update on the development of synthetic methods and pharmacological diversification. *Bioorganic & Medicinal Chemistry*. 2016; 24(11):2361-2381
28. Li D, Ambrogio L, Shimamura T, et al. BIBW2992, an irreversible EGFR/HER2 inhibitor highly effective in preclinical lung cancer models. *Oncogene*. 2008;27(34):4702-4711.

Graphical Abstract



1a

EGFR IC_{50} = 0.7 nM
Her2 IC_{50} = 39.2nM

1d

EGFR IC_{50} = 0.6 nM
Her2 IC_{50} = 42.2nM

1v

EGFR IC_{50} = 1.4 nM
Her2 IC_{50} = 10.9nM

Highlights

- New quinazoline derivatives as dual EGFR/HER-2 inhibitors
- *In vitro* evaluation of anti-proliferation activity of twenty four compounds on NCI H1975, HCC 827, A431, MDA-MB-453 cell lines.
- Identified dual EGFR/HER-2 inhibitors in nanomolar (nM) concentration.
- Compounds **1a**, **1d** and **1v** were found better EGFR/HER-2 inhibitors than afatinib.
- *In vivo* efficacy of **1d** was better than afatinib on nude mice NCI H1975 xenograft tumor model.