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Synthesis and biological evaluation of 2,4-diaminopyrimidine-5-carbonitrile and N-(2-amino-5-cyanopyrimidin-4-yl)benzamide derivatives as EGFR inhibitors

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Abstract: A series of 2,4-diaminopyrimidine-5-carbonitrile and N-(2-amino-5-cyanopyrimidin-4-yl) benzamide derivatives (**5–14**) were synthesized and their chemical structures were confirmed by 1 H, 13C NMR and mass spectral data. Anticancer activity of all the synthesized compounds were evaluated for in vitro cytotoxic activity against a panel of four human cancer cell lines i.e., human breast (MCF-7,), cervical cancer (C33A), oral (KB) and prostrate (DU-145). All the examined compounds, demonstrated potent to moderate anticancer activity. Among all the synthesized compounds, **6** and **11** were exhibited more potent activity. Docking studies for **6** and **11** into **EGFR active site** was carried out to investigate their potential binding modes. Therefore, compounds **6** and **11** can be considered as fascinating candidates for further expansion of more potent anticancer agents.

Keywords: Anticancer, EGFR inhibitors, Molecular docking, 2,4-diaminopyrimidine-5-carbonitrile and N-(2-amino-5-cyanopyrimidin-4-yl) benzamide.

Cancer is a foremost health problem worldwide and is the leading source of human mortality exceeded only by cardiovascular diseases [1-3]. This fact makes the innovation of new anticancer drugs and more effective treatment strategies for cancer of utmost importance

as traditionally prescribed chemotherapeutic agents have problems with toxicity and drug resistance. We are at an important crossroad in cancer research and clinical oncology where we should believe courageous new strategies for cancer treatment [4-5]. Immense progress have been made mapping out the cellular pathways altered in tumors and the pathways that counter to cancer treatment. The obvious significance of the components of DNA damage response pathways as potential cancer therapeutic targets has stimulated researchers and pharmaceutical companies to build up numerous chemical inhibitors for many of the proteins involved in these pathways [6-7]. Even though, various kinase inhibitors have been discovered in recent times and several have been successfully developed for treatment of cancer including Gleevec, Iressa, Tarceva, Tykerb, and Sutent, still there is strong required for breakthrough of improved cytotoxic agents[8-9]. As most of the solid human cancer tumor are multi causal in nature and their treatment with "mechanism-based" agent alone is unlikely to be successful, so a combination of these inhibitors with a better cytotoxic drug is likely to be a good strategy. A receptor tyrosine kinase, epidermal growth factor receptor (EGFR), demonstrates a critical function regarding the regulation of several cellular roles such as cell survival, proliferation, differentiation and migration [9]. EGFR mediates intracellular signaling (intrinsic intracellular protein-tyrosine kinase activity) is reply to different extracellular stimuli (endogenous ligand, like epidermal growth factor (EGF) and transforming growth factor α (TGFα)), foremost to DNA synthesis and cell growth [10,11]. EGFR over expression caused by Mutations which and establishment are associated with wide variety of cancer types as breast cancer, colorectal carcinoma, non-small cell lung cancer and pancreatic cancer [12]. Consequently, disruption of the signaling pathway of EGFR, either extracellular by blocking the binding site of EGFR inhibiting the tyrosine kinase activity, is significant in cancer prevention and treatment [13,14]. It has been recognized that EGFR is one of the most significant targets for improvement of novel cancer therapeutics [15–17]. Based on the above information, require and huge interest in the discovery of new chemical entities and novel lead structures. Quinazoline is one of the most vital heterocyclic scaffolds that become known as a potential privileged scaffold in cancer drug discovery [18–20]. Interestingly, there are numerous clinically accepted quinazoline-based anticancer drugs with potent EGFR-TK inhibitory activity such as Gefitinib [21], Afatinib [22], Erlotinib [23], Icotinib [24] and Lapatinib (Fig. 1) [25].

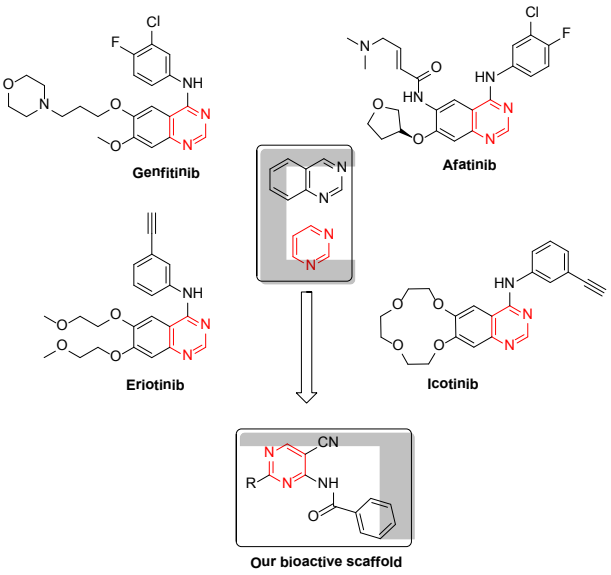


Fig. 1 An amount of clinically approved EGFR-TK inhibitors based on quinazoline and quinoline scaffolds.

The synthesis of 2,4-diaminopyrimidine-5-carbonitrile and N-(2-amino-5-cyanopyrimidin-4-yl)benzamide that are known to inhibit EGFR-TK, phosphodiesterase and ATPase, AMPA, Gly/NMDA and Kainate receptor, xanthine oxidase and benzodiazepine receptor. Conventionally, 2,4-diaminopyrimidine-5-carbonitrile are synthesized in a multi-step fashion. Design and assembly of compound

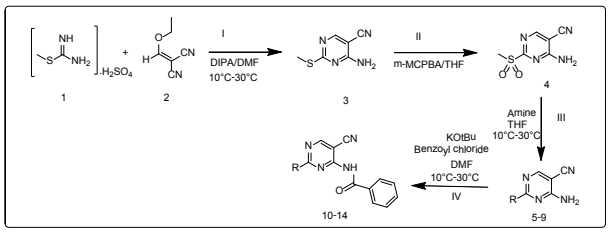
collections based on small molecules generated from multicomponent routes, play a decisive role in driving drug discovery research, since their protein targets exhibit connectivity distribution closer to human disease genes. Owing to their structural resemblance with known anticancer drug, erlotinib and gefitinib, biological investigation of 2,4-diaminopyrimidine-5-carbonitrile on cancer cell lines of human breast (MCF-7,), cervical cancer (C33A), oral (KB) and prostrate (DU-145) was carried out. A corresponding focused compound collection produced mechanism based inhibitors of Epidermal Growth Factor Receptor (EGFR) cancer cell deaths.

Experimental

All reagents and solvents were purchased from commercial sources and used without purification. NMR spectra were recorded with 200, 300, 400 MHz spectrometers for ^{1}H NMR and 50, 75, 100 MHz for ^{13}C NMR on Bruker Supercon Magnet Avance DRX-300 spectrometers in deuterated solvents with TMS as internal reference (chemical shifts δ in ppm, coupling constant *J* in Hz.). Multiplicities are reported as follows: singlet (s), doublet (d), triplet (t), multiplet (m), and broad singlet (br s). Mass spectra and HRMS were taken in the ESI positive ion mode. Microwave reactions were conducted using a Biotage Initiator in 10-mL glass tubes, sealed with Teflon septum and placed in the microwave cavity. The reaction progress was monitored by thin layer chromatography (TLC) on pre-coated silica gel plates. Column chromatography was performed over Merck silica gel (230-400 flash). All compounds were characterized by TLC, ^{1}H NMR and ^{13}C NMR, MS and HRMS.

General Procedure for the synthesis of various 2,4-diaminopyrimidine-5-carbonitrile and N-(2-amino-5-cyanopyrimidin-4-yl) benzamide derivatives

The synthesis of Synthesis and biological evaluation of 2,4-diaminopyrimidine-5-carbonitrile and N-(2-amino-5-cyanopyrimidin-4-yl)benzamide derivatives as EGFR inhibitors involved following steps:



Reagent and condition: (I) diisopropylethyl amine, DMF, 10°C to 25-30°C.; (II) 3-chloro peroxybenzoic acid, THF, 10°C to 25-30°C.;; (III) amines, THF, reflux; (IV) benzoyl chloride, potassium tert. butoxide, DMF, 10°C to 25-30°C

Compound 4-amino-5-cyano-2-methylthiopyrimidine (5-9) was synthesize by the reaction of S-methyl thiourea hydrogen sulphate and ethoxymethylene malonitrile (EMMN) in the presence of diisopropylethylamine in dry DMF at 0°C to r.t. Compound 3 was oxidized with m-chloroperoxybenzoic acid to obtain the sulphone (4), which on nucleophilic substitution with various amines furnished the compounds 5-9. Compounds 5-9 were benzoylated separately with benzoyl chloride in the presence of potassium tert. butoxide in dry DMF at 0° C–r.t. to obtain compounds 10–14.

Characterization data of compounds:

Compound 5: 4-amino-2-morpholinopyrimidine-5-carbonitrile

Solid, Yield 73%, ^{1}H NMR (400 MHz, $CDCl_3$): δ 12.43 (s, 1H), 8.78 (s, 1H), 5.98 (s, 2H), 4.56 – 4.36 (m, 4H), 4.34 – 4.23 (m, 4H)., ^{13}C NMR (101 MHz, CDCl3) δ 162.96, 161.26, 160.97, 133.37, 128.99, 116.94, 79.48, 66.71, 45.87, 44.13 ppm, HRMS (ESI) Calcd. for $C_9H_{11}N_5O$ $[M+H]^+$ 206.1036 Found 206.1001.

Table 1. Synthesis of substituted 4-amino-5-cyano-2-methylthiopyrimidine system

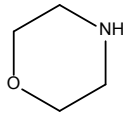
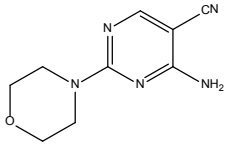
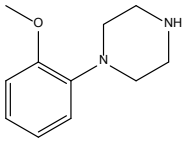
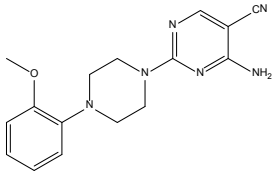
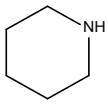
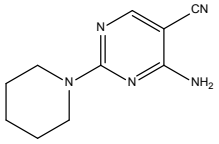
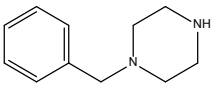
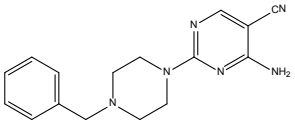
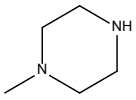
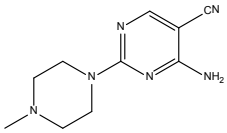
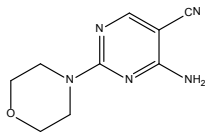
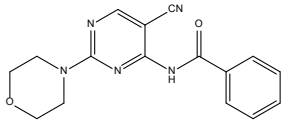
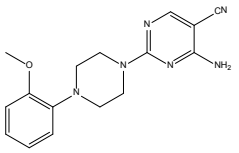
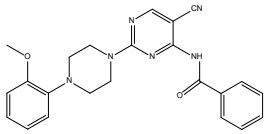
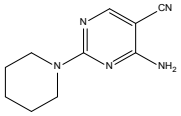
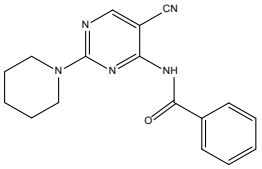
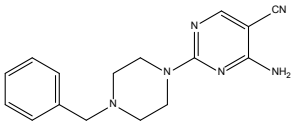
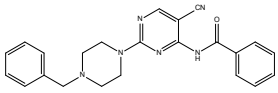
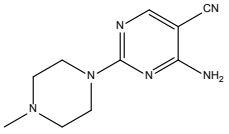
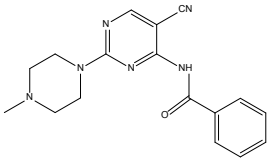
entry	amine	product	Yield
1		 **5**	**73%**
2		 **6**	**67%**
3		 **7**	**57%**
4		 **8**	**49%**
5		 9	**54%**

Table 2. Synthesis of substituted N-(2-amino-5-cyanopyrimidin-4-yl)benzamide derivatives system

entry	amine	product 15	Yield[a]
1		 **10**	**54%**
2		 **11**	**67%**
3		 **12**	**55%**
4		 **13**	**49%**
5		 **14**	**54%**

Compound 6: 4-amino-2-(4-(2-methoxyphenyl)piperazin-1-yl)pyrimidine-5-carbonitrile

Solid, Yield 67%, ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.61 (s, 1H), 7.46 – 7.37 (m, 1H), 7.35 – 7.23 (m, 3H), 5.67 (s, 2H), 4.48 – 4.34 (m, 4H), 4.27 (s, 3H), 3.76 – 3.27 (m, 4H). ppm, ^{13}C NMR (101 MHz, CDCl3) δ 163.09, 161.43, 160.99, 152.35, 123.62, 121.12, 118.51, 117.18, 111.32, 79.21, 77.48, 55.55, 50.73, 44.03,, HRMS (ESI) Calcd. for $C_{16}H_{18}N_6O$ $[M+H]^+$ 311.1615 Found 311.1611

Compound 7: 4-amino-2-(piperidin-1-yl) pyrimidine-5-carbonitrile

Solid, Yield 57%, ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.82 (s, 1H), 5.91 (s, 2H), 4.55 – 4.24 (m, 4H), 2.36 – 2.24 (m, 2H), 2.24 – 2.12 (m, 4H), 2.07 – 1.97 (m, 2H)., ^{13}C NMR (101 MHz, CDCl3) δ 163.02, 161.26, 160.65, 133.46, 130.14, 128.51, 117.38, 78.48, 77.16, 45.94, 45.02, 25.90, 24.75., HRMS (ESI) Calcd. for $C_{10}H_{13}N_5$ $[M+H]^+$ 204.1244 Found 204.1243

Compound 8: 4-amino-2-(4-benzylpiperazin-1-yl)pyrimidine-5-carbonitrile

Solid, Yield 49%, ^{1}H NMR (400 MHz, CDCl3) δ 8.82 (s, 1H), 7.99 – 7.93 (m, 4H), 7.92 – 7.88 (m, 1H), 5.86 (s, 2H), 4.57 – 4.39 (m, 4H), 4.16 (s, 2H), 3.17 – 3.01 (m, 4H), ^{13}C NMR (101 MHz, CDCl3) δ 163.05, 161.39, 160.93, 137.74, 129.32, 128.45, 127.39, 117.21, 79.08, 76.84, 63.11, 52.91, 43.79,, HRMS (ESI) Calcd. for $C_{16}H_{18}N_6$ $[M+H]^+$ 295.1666 Found 295.1659.

Compound 9: 4-amino-2-(4-methylpiperazin-1-yl)pyrimidine-5-carbonitrile

Solid, Yield 54%, ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.19 (s, 1H), 5.32 (s, 2H), 4.26 – 3.14 (m, 4H), 2.56 – 2.36 (m, 4H), 2.30 (s, 3H).ppm, ^{13}C NMR (101 MHz, CDCl3) δ 163.07, 161.38, 160.97, 117.16, 79.18, 54.85, 46.20, 43.68., HRMS (ESI) Calcd. for $C_{10}H_{14}N_6$ $[M+H]^+$ 219.1353 Found 219.1353.

.

Compound 10: N-(5-cyano-2-morpholinopyrimidin-4-yl)benzamide

Solid, Yield 54%, ^{1}H NMR (400 MHz, $CDCl_3$): 8.78 (s, 1H), 8.42-8.33 (m, 2H), 8.20-8.07 (m, 3H), 6.27 (br s, 1H) 4.63 – 4.42 (m, 4H), 4.42 – 4.37 (m, 4H)., ^{13}C NMR (101 MHz, CDCl3) δ 168.3 162.96, 161.26, 160.97, 134.6, 133.37, 132.4, 128.99, 128.6, 126.9, 116.94, 79.48, 66.71, 45.87, 44.13 ppm, HRMS (ESI) Calcd. for $C_{16}H_{15}N_5O_2$ $[M+H]^+$ 310.1299 Found 309.1200

Compound 11: N-(5-cyano-2-(4-(2-methoxyphenyl)piperazin-1-yl)pyrimidin-4-yl)benzamide

Solid, Yield 67%, ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.61 (s, 1H), 8.39-8.33 (m, 2H), 8.12-8.02 (m, 3H), 7.46 – 7.37 (m, 1H), 7.35 – 7.23 (m, 3H), 6.34 (br s, 1H), 4.48 – 4.34 (m, 4H), 4.27 (s, 3H), 3.76 – 3.27 (m, 4H). ppm, ^{13}C NMR (101 MHz, CDCl3) δ 169.2,164.9, 160.7, 159.2, 151.35, 133.6, 131.9,127.9,126.4,123.40, 120.12, 117.99, 116.18, 110.42, 79.66, 76.48, 56.54, 50.90, 43.77,, HRMS (ESI) Calcd. for $C_{23}H_{22}N_6O_2$ $[M+H]^+$ 415.1877 Found 415.1789

Compound 12: N-(5-cyano-2-(piperidin-1-yl) pyrimidin-4-yl)benzamide

Solid, Yield 55%, ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.88 (s, 1H), 8.65-8.50 (m, 2H), 8.20-8.09 (m, 3H), 6.37 (br s, 1H), 4.65 – 4.57 (m, 4H), 2.77 – 2.67 (m, 2H), 2.34 – 2.22 (m, 4H), 2.17 – 1.99 (m, 2H)., ^{13}C NMR (101 MHz, CDCl3) δ 168.7 163.7, 161.66, 160.99, 134.6 133.46, 131.4, 130.14, 128.7, 128.51, 126.9,117.88, 78.78, 77.16, 46.54, 45.55, 26.30, 24.95., HRMS (ESI) Calcd. for $C_{17}H_{17}N_5O$ $[M+H]^+$ 308.1506 Found 308.1504

Compound 13: N-(2-(4-benzylpiperazin-1-yl)-5-cyanopyrimidin-4-yl)benzamide

Solid, Yield 49%, ^{1}H NMR (400 MHz, CDCl3) δ 9.01 (s, 1H), 8.67-8.53 (m, 2H), 8.33-8.21 (m, 3H), 8.07-8.00 (m, 4H), 7.98 – 7.93 (m, 1H), 6.69(br s, 1H), 4.63 – 4.42 (m, 4H), 4.23(s, 2H),

3.25 – 3.07 (m, 4H), ^{13}C NMR (101 MHz, CDCl3) δ 169.2, 164.75, 162.59, 161.25, 138.21,135.2, 134.9, 129.62, 128.9, 128.55, 127.72, 117.9, 79.75, 76.82, 63.69, 53.98, 44.79,, HRMS (ESI) Calcd. for $C_{23}H_{22}N_6O$ $[M+H]^+$ 399.1928 Found 399.1917.

Compound 14: N-(5-cyano-2-(4-methylpiperazin-1-yl)pyrimidin-4-yl) benzamide

Solid, Yield 54%, ^{1}H NMR (400 MHz, $CDCl_3$): δ 8.24 (s, 1H), 8.11-8.03 (m, 2H), 8.01-7.91 (m, 3H), 6.21(br s, 1H), 4.21 – 3414 (m, 4H), 2.67 – 2.4 (m, 4H), 2.50 (s, 3H).ppm, ^{13}C NMR (101 MHz, CDCl3) δ 168.1, 163.07, 162.38, 161.94, 134.0, 131.4, 128.5, 127.0, 117.55, 79.34, 55.85, 47.80, 43.78., HRMS (ESI) Calcd. for $C_{17}H_{18}N_6O$ $[M+H]^+$ 323.1615 Found 323.1550

Anticancer activity

The anticancer activity of the synthesized compounds was evaluated against four cancerous cell lines; human breast (MCF-7,), cervical cancer (C33A), oral (KB) and prostrate (DU-145) using (SRB) colorimetric assay. Doxorubicin and Erlotinib were included in the experiments as reference cytotoxic compounds for all the tested cell lines. The results were expressed as median growth inhibitory concentration (IC_{50}) values, which represent the concentration of a drug that is required for 50% inhibition of cell growth after 48 h of incubation, compared to untreated controls (Table 2).

Table 2. *In vitro* anticancer activity of compounds (**5-14**).

Compound No.	IC_{50} (μg/mL)				
	DU 145	MCF7	C33A	KB	VERO
5	15.8	23.4	21.7	15.6	24.3
6	3.0	1.4	1.6	2.8	37.8
7	20.2	28.0	24.9	21.6	33.7
8	8.9	4.3	3.3	3.9	8.2
9	10.1	5.2	4.2	4.5	9.8
10	2.5	3.2	3.1	6.0	2.6
11	0.2	1.6	4.2	1.5	5.9
12	4.5	3.9	2.5	5.1	7.8
13	21.3	35.2	26	33.4	29.7
14	23.0	32.4	19.8	22.8	37.8

All of the tested compounds showed potent to moderate activity with IC_{50} values ranging from 0.21 to 35.2 μM. In particular, compounds 6 and 11 were the most active compounds through this study with IC_{50} values equal 1.4and 0.2 μM, respectively.

Molecular docking

Docking of the most potent EGFR-TK inhibitors (6 and 11) was passed out to study their pattern of binding and potential binding interactions into the ATP binding site of the EGFR kinase domain. The ability of compounds 6 and 11 to interact with the key amino acids in the ATP binding site of EGFR-TK rationalized their promising antitumor activities. *In silico* study clearly depicted that compound 11 poses higher and strong binding ability with EGFR (PDB ID: 1XKK) over compound 6. Compound 11 and EGFR bind with binding energy -8.25 Kcal/Mol, dissociation constant (Ki) 898.49 nM and N20 atom of compound 11 formed hydrogen bond with Nitogen atom of GLY796 with distance of 3.03318 Å, total 19 amino acids of EGFR are involved in hydrophobic interactions. On other hand side Compound 6 posses lesser binding energy than compound 11, it was -6.28 Kcal/Mol, 24.81 μM Ki and also form a hydrogen bond with H41 atom of compound 6 with Oxygen atom of Leu788 residue of EGFR hydrogen bond distance is 2.24605 Å, at last total 16 amino acids of EGFR are involved in hydrophobic interactions with compound 6 (as shown in table 3 and Figure 2).

Table3: Binding pattern of Compound 6 and 11 with EGFR protein.

S. No.	Receptor	Ligand	Binding Energy (Kcal/Mol)	Ki	Binding Residues	H-Bond	Distance of H-Bond (Å)
1	1XKK	Compound 6	-6.28	24.81 µM	Leu718,Val726,Ala743 Ile744,Lys745,Met766 Leu777,Leu788,Ile789 Thr790,Gly796,Cys797 Arg841,Leu844,Thr854 Met1002	:Compound 6:H41 - A:LEU788:O	2.24605
2	1XKK	Compound 11	-8.25	898.49 nM	Leu718,Val726,Ala743 Lys745,Met766,Leu777 Leu788,Ile789,Thr790 Leu792,Met793,Gly796 Cys797,Asp800,Leu844 Thr854,Asp855,Leu858 Met1002	A:GLY796:N - :Compound 11:N20	3.03318

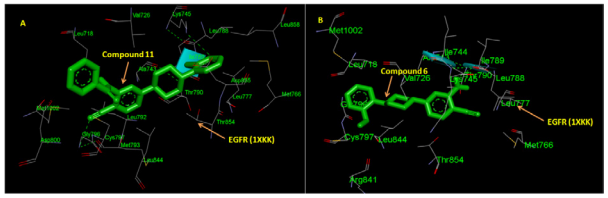


Figure 2: A) Binding pose of Compound 11 and EGFR B) Binding pose of Compound 6 and EGFR.

Material and Methods:

RCSB protein data bank was used to procure 3D structure of EGFR (PDB ID 1XKK: Chain A). 1XKK was edited to remove HETATM using Discovery Studio Visualizer 3.1. Auto Dock Tool 4 (MGL Tool) was used for the molecular docking, identification of binding affinities and poses of ligands and proteins. (Morris GM *et al* 2009 and Dhasmana A *et al* 2016).

Conclusions

In summary, a series of novel 2,4-diaminopyrimidine-5-carbonitrile and N-(2-amino-5-cyanopyrimidin-4-yl)benzamide derivatives d was designed, synthesized and evaluated as potent EGFR inhibitors. The results showed that most of synthesized compounds exhibited moderate to high anticancer activities against four human tumor cell lines including human breast (MCF-7,), cervical cancer (C33A), oral (KB) and prostrate (DU-145) using (SRB) colorimetric assay and ***EGFR Kinase***.

References

1. A. Jemal, M.M.Center, C. DeSantis, E. M Ward. **2010**, 19, 1893–1907.
2. M. Rashid, A. Husain, R. Mishra, Eur J Med Chem. **2012**, 54, 855–866.
3. F. Rojo, J.Albanell, A. Rovira, J.M. Corominas, F. Manzarbeitia. Semin Diagn Pathol. **2008**, 25, 245–261.
4. B.W. Stewart, C.P. Wild, World Cancer Report 2014, World Heal. Organ., **2014**.
5. A. T. Lindsey, B. Freddie, R. L. Siegel, J. Ferlay, A. Jemal, Global Cancer Statistics, 2012, CA: Cancer J. Clin. **2015**, 65 (2) 87–108.
6. M.J. O'Connor, Mol. Cell **2015**, 60, 547–560.
7. C. Yewale, D. Baradia, I. Vhora, S. Patil, A. Misra, Biomaterials, **2013**, 34, 8690–8707.
8. N.R. Patel, B.S. Pattni, A.H. Abouzeid, V.P. Torchilin, Adv. Drug Deliv. Rev., **2013**, 65 (13–14) , 1748–1762.
9. P.D.W. Eckford, F.J. Sharom, Chem. Rev. **2009**, 109, 2989–3011.
10. C. Yewale, D. Baradia, I. Vhora, S. Patil, A. Misra, Biomaterials, **2013**, 34, 8690–8707.
11. M. Katz, I. Amit, Y. Yarden, Biochim. Biophys. Acta - Mol. Cell Res. **2007**, 1773, 1161–1176.
12. N. Prenzel, O.M. Fischer, S. Streit, S. Hart, A. Ullrich,

Endocr. Relat. Cancer, **2001**, 8, 11–31.
13. R. Roskoski, Biochem. Biophys. Res. Commun., **2004**, 319, 1–11.
14. N. Tebbutt, M.W. Pedersen, T.G. Johns, Nat. Rev. Cancer, **2013**, 13, 663–673.
15. J. Zhang, P.L. Yang, N.S. Gray, Nat. Rev. Cancer **2009**, 9, 28–39.
16. H. Masuda, D. Zhang, C. Bartholomeusz, H. Doihara, G.N. Hortobagyi, N.T. Ueno, Breast Cancer Res. Treat. **2012**, 136, 331 -345.
17. R. Costa, Cancer Treat. Rev. **2017**, 53, 111–119.
18. R.S.M. Ismail, Eur. J. Med. Chem., **2018**, 155, 782–796.
19. S. Shagufta, I. Ahmad, Medchemcomm , **2017**, 5, 871–885.
20. D. Das, J. Hong, Eur. J. Med. Chem., **2019**, 170, 55–72.
21. R.S.M. Ismail, N.S.M. Ismail, S. Abuserii, D.A. Abou El Ella, Futur. J. Pharm. Sci. **2016**, 2, 9–19.
22. A.J. Barker, Bioorg. Med. Chem. Lett. **2001**, 11, 1911–1914.
23. R.T. Dungo, G.M. Keating, **2013**, 73, 1503–1515.
24. Y. Wang, G. Schmid-Bindert, C. Zhou, Ther. Adv. Med. Oncol., **2012**, 4,19–29.
25. F. Tan, et al., Lung Cancer , **2012**, 76, 177–182.
26. J.E. Frampton, Lapatinib, Drugs , 2009, 69, 2125–2148.