Bioorganic & Medicinal Chemistry Letters 28 (2018) 2328–2337

Contents lists available at ScienceDirect

Bioorganic & Medicinal Chemistry Letters

journal homepage: www.elsevier.com/locate/bmcl

Synthesis of novel hetero ring fused pyridine derivatives; Their anticancer activity, CoMFA and CoMSIA studies

G. Santhosh Kumar [a], Y. Poornachandra [b], Shravan Kumar Gunda [c], K. Ratnakar Reddy [a], Jaheer Mohmed [c], Kamal Shaik [d], C. Ganesh Kumar [b], B. Narsaiah [a,*]

[a] Fluoro Organic Division, CSIR-Indian Institute of Chemical Technology, Tarnaka, Hyderabad 500007, India
[b] Medicinal Chemistry and Pharmacology Division, CSIR-Indian Institute of Chemical Technology, Tarnaka, Hyderabad 500007, India
[c] Bioinformatics Division, PGRRCDE, Osmania University, Hyderabad 500007, India
[d] Department of Chemical Engineering, Visvesvaraya National Institute of Technology, Nagpur 440010, India

ARTICLE INFO

Article history:
Received 10 February 2018
Revised 8 April 2018
Accepted 11 April 2018
Available online 21 April 2018

Keywords:
Carbohydrazide Schiff's bases
Pyrido[3′,2′:4,5]furo[3,2-*d*]pyrimidin-4(3*H*)-one
Anti-cancer activity
3D QSAR
CoMFA and CoMSIA studies

ABSTRACT

A series of novel furo[2,3-*b*]pyridine-2-carboxamide **4a**–**h**/pyrido[3′,2′:4,5]furo[3,2-*d*] pyrimidin-4(3*H*)-one derivatives **5a**–**p** were prepared from pyridin 2(1*H*) one **1** via selective O-alkylation with α-bromoethylester followed by cyclization, then reaction with different aliphatic primary amines to obtain **4** and further reaction with triethyl orthoacetate/triethyl orthoformate. Also prepared novel furo[2,3-*b*]pyridine-2-carbohydrazide Schiff's bases **7a**–**h** and pyrido [3′,2′:4,5]furo[3,2-*d*]pyrimidin-4(3*H*)-one derivatives **8a**–**h** starting from furo[2,3-*b*]pyridine carboxylate derivatives **3** by reaction with hydrazine hydrate to form **6** and reaction with diverse substituted aldehydes and cyclization. Products **4a**–**h**, **5a**–**p**, **7a**–**h** and **8a**–**h** were screened against four human cancer cell lines (HeLa, COLO205, Hep G2 and MCF 7) and one normal cell line (HEK 293). Compounds **4e**, **4f**, **4g**, **5h**, **7c**, **7d**, **7e** and **7f** showed significant anticancer activity against all the cell lines at micro molar concentration and found to be non-toxic to normal cell line. Studies for HeLa, COLO205 and MCF-7 using CoMFA and CoMSIA. Models from 3D-QSAR provided a strong basis for future rational design of more active and selective HeLa, COLO205 and MCF-7 cell line inhibitors.

Present statistics indicate that the cancer disease is second common cause of death after heart disease and continuously growing as worldwide killer.[1] Although chemotherapy is the backbone for cancer treatment, the use of chemotherapeutics is often limited due to undesirable side effects. Therefore, identification of new agents as targets for the treatment of cancer is very important. Some of the furopyridine and pyrimidi none derivatives (5,6 fused ring systems) found to have biological and chemotherapeutic importance. Furopyridines are structural analogues to indoles, pyrrolopyridines which are frequently employed as core scaffolds and play a significant role in promoting activity.[2,3] Furo[2,3-*b*]pyridine skeleton is rarely found in naturally occurring alkaloids;[4] however, it showed activity against HIV,[5] CNS disorders,[6] skin diseases[7] and hyperglycemia.[8] Furo[2,3-*b*]pyridine derivatives also demonstrated in vitro activity for tubulin polymerization,[9] Lck[10] and Akt[11] kinase inhibitors. A specific drug Cicletanine, L-754, 394 based on furopyridine, shows an antihypertensive with vasorelaxant, diuretic property[12,13] and potent HIV protease inhibitor. Fluorouracil and Tegafur based on pyrimidinone skeleton used as anticancer agents are outlined in Fig. 1.

Fused pyrimidines have a long and distinguished history extending from the days of their discovery as important constituents of nucleic acids to their current use in the chemotherapy. Hydrazones have been demonstrated to possess antimicrobial, anticonvulsant, analgesic, anti-inflammatory, anti-platelet, antitubercular and anti-tumor activities. Hydrazide derivatives are not only intermediates, coupling products can be synthesized by using the active hydrogen component of —CONHN=CH— azomethine group.[14] Similarly, thieno[2,3-*b*]quinoline-2-carboxamide derivatives are considered as anticancer agents.[15] Further, it was found that the fluorine[16] or trifluoromethyl[17,18] group at a strategic position of an organic molecule dramatically alters the properties of molecule in terms of lipid solubility, oxidative thermal stability thereby enhances the transport mechanism and bio-efficacy. However, no reports are available on synthesis of fluorinated furo [2,3-*b*] pyridine derivatives except our reports.[19,20] In continuation of our efforts, we designed and synthesized a series of novel trifluoromethyl substituted furo [2,3-*b*] pyridine-2-carbohydrazide Schiff's bases and pyrido [3′,2′:4,5] furo [3,2-*d*] pyrimidin-4(3*H*)-

* Corresponding author.
E-mail address: narsaiah@iict.res.in (B. Narsaiah).

https://doi.org/10.1016/j.bmcl.2018.04.031
0960-894X/

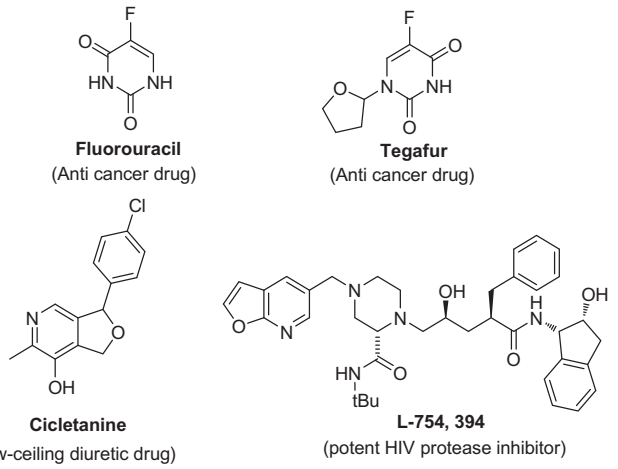

Fig. 1. Bio-active compounds based on furopyridine and pyrimidinone scaffolds.

one derivatives and screened them for anticancer activity against four human cancer cell lines. Compounds **4e**, **4f**, **4g**, **5h**, **7c**, **7d**, **7l** and **7o** which showed promising anticancer activity have been identified.

The 3-cyano-4-trifluoromethyl-6-substituted pyridine 2(1*H*) one **1** was reacted with 2-bromoethyl acetate under basic conditions and obtained selectively 2-O-ethylacetoxy-3-cyano-4-trifluoro methyl-6-substituted pyridine derivatives **2**. Compounds **2** were cyclized in DMF using potassium carbonate as base and obtained furo[2,3-*b*]pyridine derivatives **3**. The reaction sequence includes selective O-alkylation then abstraction of proton by base from an active methylene followed by cyclization onto nitrile carbon. This type of cyclization is also known as Thorpe-Ziegler cyclization. Compounds **3** were reacted with different substituted aliphatic primary amines to result in carboxamide derivatives **4** and were further independently reacted with triethyl orthoacetate and triethyl orthoformate to obtain pyrido furo pyrimidinone derivatives **5a**–**p**. The sequence of reactions outlined in Scheme 1.

Compounds **3** were reacted with hydrazine hydrate to result carbohydrazide derivatives **6** which were further reacted with diverse substituted aromatic aldehydes to obtain furo[2,3-*b*]pyridine-2-carbohydrazide Schiff's bases **7**. Compounds **7** were reacted with TEOF (triethyl orthoformate) in the presence of catalytic amount of acetic acid and obtained the final products of furo pyrido pyrimidinone derivatives **8a**–**h**. The sequence of reactions outlined in Scheme 2.

Anticancer activity & structure-activity relationship

Compounds **4a**–**h** and **5a**–**p** were screened against four human cancer cell lines such as HeLa (cervical cancer, CCL-2), COLO-205 (colon cancer, CCL-222), HepG2 (liver cancer, HB-8065) and MCF7 (breast cancer, HTB-22) using MTT assay.[21] Among all the compounds screened, compounds **4e**, **4f**, **4g**, and **5h** showed promising activity at micro molar concentration. The structure–activity relationship studies revealed that the carboxamide derivatives **4a**–**h** showed more promising activity as compared to pyrimidinone derivatives **5a**–**p**. It was attributed to the presence of amide and primary amine group in compounds **4a**–**h** and was considered as polar groups. Among all the compounds screened in **4a**–**h** series, compounds **4e**–**g** showed promising activity. Similarly, in **5a**–**p** series, the presence of thiophenyl group in 6th position was crucial for activity and compound **5h** exhibited the promising activity. The activity data is shown in Tables 1 and 2.

Compounds **7a**–**h** and **8a**–**h** were also screened against four human cancer cell lines such as HeLa, COLO-205, HepG2 and MCF7. Compounds **7a**–**h** are uncyclized Schiff's base products, while compounds **8a**–**h** were cyclized pyrimidinone products. The structure verses activity revealed that the presence of —NH_2 on furyl and amide linkage (—NHCO) are crucial for activity. Among all the compounds screened, compounds **7a**–**h** showed promising activity as compared to compounds **8a**–**h**. Compounds **7c**, **7d**, **7e** and **7f** were found to exhibit high activity. Among all the compounds **7c**, **7d**, **7e** and **7f** were found to be more potent with IC_{50} values of <10 μg/mL on all the tested cancer cell lines. The activity data to this regard is tabulated in Tables 3 and 4. Compounds **7c** and **7d** were considered as lead molecules for further optimization. The introduction of thiophenyl group at 6th position of furopyridine and chloro, bromo substituents at 4th position of phenyl ring enhanced the activity to a greater extent as compared to other compounds. However, the same substituents for compounds **8c** and **8d** could not show activity due to non-availability of —NH_2 group and uncyclized amide linkage (—NHCO). Remaining compounds were not active up to the concentration of 60 μg/mL. The activity data is shown in Table 3.

All the above compounds were also screened against human normal cell line (HEK-293, Human Embryonic Kidney cells, CRL-1573) and they were found to be not cytotoxic up to the concentration of 51 μg/mL to 124 μg/mL as compared to 5-Fluorouracil (19.6 μg/mL).

QSAR introduction

In general, the 3D-QSAR techniques are valuable methods of ligand-based drug design by correlating physicochemical properties from a set of related compounds to their known molecular property or molecular activity values. Validation of QSAR models plays the vital role in defining the applicability of the QSAR model for the prediction of designed molecules. QSAR model is mostly used to correlate properties, *i.e.* biological activities with chemical structures, and also used to predict the biological activity of non-synthesized compounds, which are structurally related to training sets. The present investigation reports the first application of 3D-QSAR to study of furo [2,3-*b*] pyridine and pyrido [3′,2′:4,5] furo [3,2-*d*] pyrimidin-4(3*H*)-one derivatives as potent anticancer agents. We studied twenty-one compounds for HeLa and COLO205 cell lines, twenty-six compounds for MCF-7 cell line inhibitors as anticancer agents using CoMFA (comparative molecular field analysis)[22] and CoMSIA (comparative molecular similarity indices analysis).[23] Models obtained from 3D-QSAR studies provided a strong basis for future rational design of more active and selective HeLa, MCF-7 and COLO205 cell line inhibitors.

Results and discussion

CoMFA and CoMSIA

CoMFA and CoMSIA methods were applied to derive 3D-QSAR models for furo [2,3-*b*] pyridine and pyrido [3′,2′:4,5] furo [3,2-*d*] pyrimidin-4(3*H*)-one derivatives as potential anticancer inhibitors. The statistical results of CoMFA and CoMSIA analysis are summarized in Table 4. Best predictions were obtained with CoMFA standard model involving cross-validated coefficient (q^2) = 0.820, 0.876 and 0.853 for HeLa, MCF-7 and COLO205 cell lines, respectively, correlation coefficient (r^2) = 0.960, 0.959 and 0.978 for HeLa, MCF-7 and COLO205, respectively. Standard Error of Estimate (SEE) = 0.084, 0.081 and 0.056 for HeLa, MCF-7 and COLO205, respectively. Cross Validation (cv) = 0.818, 0.867 and 0.841 and Fischer statistic (F-value) = 47.511, 75.303 and 87.829 for HeLa,