European Journal of Medicinal Chemistry 78 (2014) 65–71

Contents lists available at ScienceDirect

European Journal of Medicinal Chemistry

journal homepage: http://www.elsevier.com/locate/ejmech

Original article

Design, synthesis and bioevaluation of *N*-trisubstituted pyrimidine derivatives as potent aurora A kinase inhibitors

Yu Luo [a,1], Yan-Qiu Deng [a,1], Jing Wang [b], Zi-Jie Long [b], Zheng-Chao Tu [c], Wei Peng [a], Ji-Quan Zhang [a], Quentin Liu [b,**], Gui Lu [a,d,*]

[a] Institute of Drug Synthesis and Pharmaceutical Process, School of Pharmaceutical Sciences, Sun Yat-sen University, Guangzhou 510006, PR China
[b] State Key Laboratory of Oncology in South China, Cancer Center, Sun Yat-sen University, Guangzhou 510060, PR China
[c] Guangzhou Institute of Biomedicine and Health, Chinese Academy of Sciences, Guangzhou 510530, PR China
[d] Institute of Human Virology, Sun Yat-sen University, Guangzhou 510080, PR China

ARTICLE INFO

Article history:
Received 29 November 2013
Received in revised form
18 February 2014
Accepted 9 March 2014
Available online 12 March 2014

Keywords:
Pyrimidine derivative
Small molecule inhibitor
Antitumor
Aurora A kinase
Synthesis

ABSTRACT

The design and synthesis of a new series of *N*-trisubstituted (at C2, C4 and C6 respectively) pyrimidine derivatives were reported, their *in vitro* structure–activity relationships vs. aurora A kinase were also discussed. Our results demonstrated that the introduction of characteristic *N*-substituted side chain at C2 of pyrimidines possessed a potent aurora A inhibitory activity, the position and the nature of the substituents on the phenyl ring of aniline side chain played key roles in cellular kinase inhibitory potency. Most tested compounds exhibited good inhibitory activities against aurora A kinase and various human tumor cell lines. Compounds **7j**, **7m**–**n** and **7p** showed strong growth–inhibitory activities in the solid CNE-2 tumor cell and selectively blocked cell-cycle progression at the G_2/M phase.

1. Introduction

The aurora family of serine/threonine protein kinase is crucial for the proper regulation of mitosis. Mammals express three aurora kinase paralogues, known as aurora A, B and C respectively. Despite significant sequence homology, the localization and functions of these aurora kinases are distinct. Aurora A localizes to centrosomes during early S phase and is involved in centrosome maturation and separation, bipolar spindle assembly, mitotic entry, and mitotic exit. Aurora B and C are chromosomal passenger proteins, although aurora C kinase is less well known [1].

For aurora A plays an important role in cell cycle progression and carcinogenesis, the development of small molecule inhibitors of aurora A as potential molecular-targeted therapeutic intervention against cancer has been pursued by many researchers. Some of these aurora kinase inhibitors have already been under Phase I/II evaluation for various cancers.

Substituted pyrimidine is an important structure presented in a number of aurora kinase inhibitors (Fig. 1) [2–15]. For example, **VX-680** has been discovered as a pan-aurora selective inhibitor [2], although the clinical research was terminated due to the safety data (QTc prolongation was observed) [3]. **ENMD-2076** selectively inhibits aurora A kinase with an IC_{50} value of 14 nM [4]. **CYC116** is an orally available aurora kinase inhibitor which is currently undergoing Phase I clinical trials [5]. **AZD1152** shows selectivity for aurora B (K_i of 0.36 nM) over aurora A (K_i of 1.37 μM) and enters Phase II clinical studies [6]. **MLN8054** has been proved to inhibit aurora A selectively, and is now under Phase I clinical trials for advanced solid tumors [7].

The chemical structure of **VX-680** suggested that the replacement of *S*-substituted side chain at C2 with *N*-substituted aniline side chain should retain its affinity for aurora kinase but significantly simplify its synthesis. **ENMD-2076** inspired that the change of styrene side chain with *N*-benzyl amine side chain might also be efficient for high inhibitory activity. Moreover, it was envisaged that further improvements in terms of both potency and physical properties could be made by adjusting the *N*-substituents at C2 and

* Corresponding author.
** Corresponding author.
E-mail addresses: liuq9@mail.sysu.edu.cn (Q. Liu), lugui@mail.sysu.edu.cn (G. Lu).
[1] These authors contributed equally to this paper.

http://dx.doi.org/10.1016/j.ejmech.2014.03.027
0223-5234/

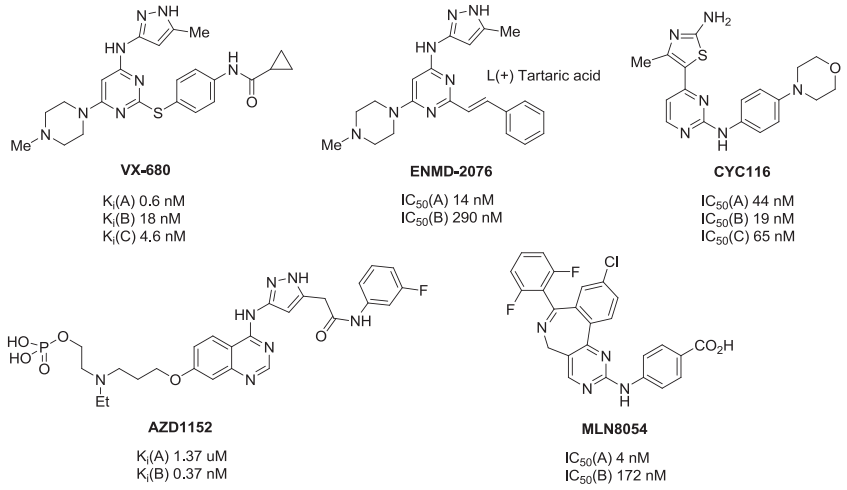


Fig. 1. Some pyrimidine-based aurora kinase inhibitors in clinical testing.

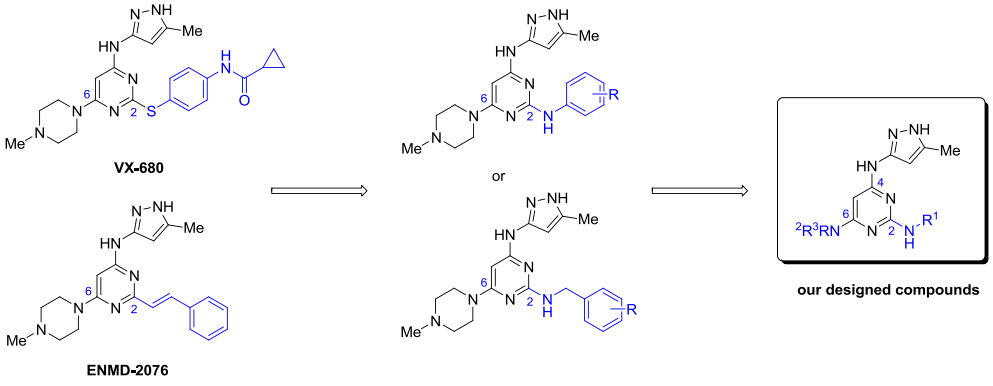


Fig. 2. Our designed *N*-trisubstituted pyrimidine derivatives.

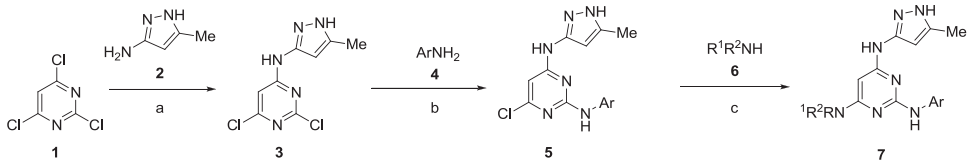
C6 of pyrimidine (Fig. 2). Herein we described the discovery and characterization of a new series of *N*-trisubstituted (at C2, C4 and C6 respectively) pyrimidine derivatives. Their *in vitro* structure–activity relationships vs. aurora A kinase and corresponding cellular potency were also analyzed.

2. Results and discussions

2.1. Chemistry

The general synthetic route for *N*-trisubstituted pyrimidine was illustrated in Scheme 1. Treatment of 2,4,6-trichloropyrimidine (**1**) with 3-amino-5-methylpyrazole (**2**) afforded C4-substituted pyrimidine (**3**) regioselectively [16,17]. Then intermediate **5** was generated via nucleophilic substitution at C2-position of pyrimidine core in the presence of various aniline derivatives [18]. Microwave assisted substitution at C6-position of pyrimidine ring gave final product **7** [19].

7e (or **7f**) was synthesized via nucleophilic substitution between **7b** and methylsulfonyl chloride (or *para*-toluensulfonyl chloride) as shown in Scheme 2. The hydrolysis of **7a** via lithium hydroxide afforded the corresponding benzoic acid **7i** (Scheme 3) [20]. 2-Benzylamine substituted pyrimidine **10** was obtained by the reaction of intermediate **3** with benzylamine in the presence of



Reagents and conditions: (a) Et_3N, EtOH, 0 °C; (b) $TsOH{\cdot}H_2O$, *n*-BuOH or 1,4-dioxane, 100-140 °C; (c) 1,4-dioxane, under microwave, 150-180 °C.

Scheme 1. General synthesis of 2,4,6-trisubstituted pyrimidine **7**.