Bioorganic & Medicinal Chemistry Letters 28 (2018) 1386–1391

Contents lists available at ScienceDirect

Bioorganic & Medicinal Chemistry Letters

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

Discovery of novel CDK inhibitors via scaffold hopping from CAN508

Liandong Jing, Yanbo Tang, Zhiyan Xiao *

Beijing Key Laboratory of Active Substance Discovery and Druggability Evaluation, Institute of Materia Medica, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing 100050, China

ARTICLE INFO

Article history:
Received 20 December 2017
Revised 17 February 2018
Accepted 27 February 2018
Available online 8 March 2018

Keywords:
Pyrazolo[3,4-*b*]pyridine
Scaffold hopping
CAN508
CDK2
CDK9
Selectivity

ABSTRACT

Cyclin-dependent kinases (CDKs) are promising drug targets for various human diseases, especially for cancers. Scaffold hopping strategy was applied on CAN508, a known selective CDK9 inhibitor, and a series of pyrazolo[3,4-*b*]pyridine compounds were synthesized and evaluated *in vitro* as CDK2 and CDK9 inhibitors. Most compounds exhibited moderate to potent inhibitory activities against both CDK2/cyclin A and CDK9/cyclin T1 systems. Among them, compound **2e** showed IC_{50} values of 0.36 μM for CDK2 and 1.8 μM for CDK9, respectively. Notably, the scaffold alteration seems to cause a shift in the selectivity profile of the inhibitors. In contrast to CAN508, compound **2k** demonstrated remarkable selectivity toward CDK2 (265-fold over CDK9). Docking studies on compound **2k** provided hints for further design of more potent and selective CDK2/CDK9 inhibitors.

Members of the cyclin-dependent kinase (CDK) family play critical roles in the regulation of a wide range of physiological processes, including cell cycle division, proliferation, apoptosis, gene transcription, and neuronal functions.[1] In the recent two decades, accumulative evidences have revealed that CDKs are promising therapeutic targets for a variety of human diseases, especially different types of cancers.[2–4] Owing to the great biological significance of CDKs, a large number of CDK inhibitors have been developed, and currently there are about twenty CDK inhibitors under different stages of clinical investigations as cancer therapeutics.[5,6] Up to now, four CDK inhibitors have been approved by the U.S. Food and Drug Administration (FDA) (Fig. 1). Flavopiridol (Alvociclib)[7] is a pan-CDK inhibitor, and was granted an orphan drug designation by FDA for the treatment of chronic lymphocytic leukemia (CLL) in 2014. Palbociclib from Pfizer[8] is the first selective CDK4/6 inhibitor reached market. Two other selective CDK4/6 inhibitors, Ribociclib from Novartis[9] and Abemaciclib from Eli Lilly[10], have also received FDA approval in the next two years. All the three selective CDK4/6 inhibitors have been designated for the treatment of HR^+ and $HER2^-$ breast cancer. The translational success of the selective CDK 4/6 inhibitors has inspired extensive research interest in developing selective CDK inhibitors. More recently, the first selective CDK9 inhibitor, Atuveciclib (BAY1143572)[11], has also entered Phase I clinical trial as an orally available cancer therapy.

Among all the CDK subtypes, CDK2 and CDK9 have been most intensely studied. CDK2 is directly involved in the control of multiple events in the cell cycle, and inactivation of its endogenous inhibitors or overexpression of CDK2 may cause various malignancies, such as lung carcinoma, melanoma, ovarian carcinoma and pancreatic carcinoma.[12] It has also been recently confirmed that inhibition of the CDK2 kinase activity led to remarkable growth inhibition in a panel of human cancer cell lines.[13] In contrast to CDK2, CDK9 participates in the regulation of gene transcription. As a subunit of positive transcription elongation factor b (P-TEFb), CDK9 phosphorylates the C-terminal domain of RNA pol II and then promote transcription elongation.[14,15] Inhibition of CDK9 results in the rapid depletion of short-lived survival proteins (e.g. Mcl-1) and oncogenes (e.g. c-MYC).[16] Furthermore, CDK9 deregulation is also associated with AIDS and other human virus-caused diseases.[16,17] Owing to their biological significance, numerous CDK2 and CDK9 inhibitors with divergent selectivity profiles have been identified.[18,19,4]

We report herein the identification of novel CDK2/9 inhibitors via scaffold hopping based on a known selective CDK9 inhibitor, CAN508 (Fig. 2). CAN508 showed an IC_{50} value of 0.35 μM against CDK9, and exhibited over 200-fold selectivity toward CDK9 over CDK2.[20] The availability of CAN508-CDK2/CDK9 complex structures could shed light on the structural basis for selectivity and make structure-based selectivity modulation possible.[21] Furthermore, CAN508 has a relatively simple scaffold with a molecule weight of only 218, therefore, it is possible to modulate the selectivity profile by chemical manipulation on the scaffold.

As illustrated in Fig. 2, CAN508 assumed similar poses in the ATP binding pockets of both CDK2 and CDK9. Interestingly, intramolecular hydrogen bonds between an azo-nitrogen and an amino group on the pyrazole ring were observed, and a pseudo

* Corresponding author.
E-mail address: xiaoz@imm.ac.cn (Z. Xiao).

https://doi.org/10.1016/j.bmcl.2018.02.054
0960-894X/

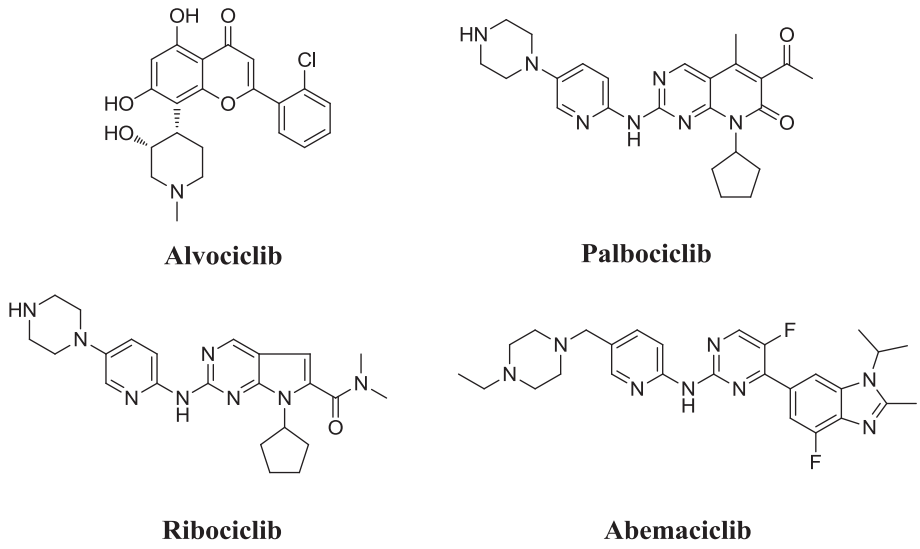

Fig. 1. CDK inhibitors approved by FDA.

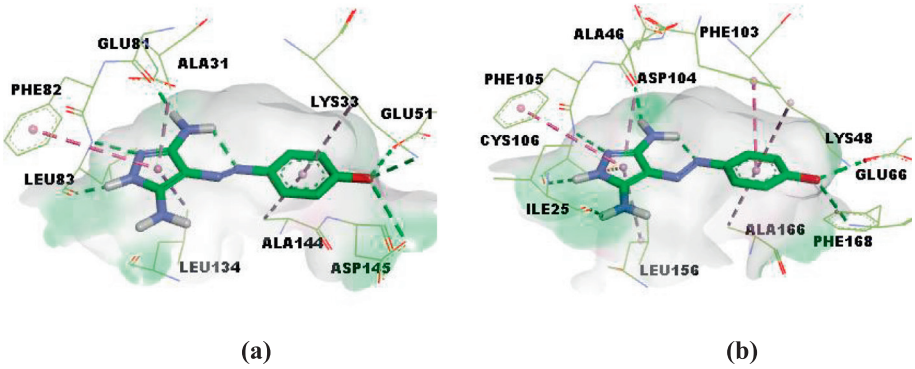

Fig. 2. Interaction modes of CAN508 with CDK2 (PDB: 3TNW)[21] (**a**) and CDK9 (PDB: 3TN8)[21] (**b**). Green dashed lines indicate hydrogen bonds, deep purple dashed lines represent cation-π or π-π stacking interactions and hydrophobic interactions are shown as light purple dashed lines.

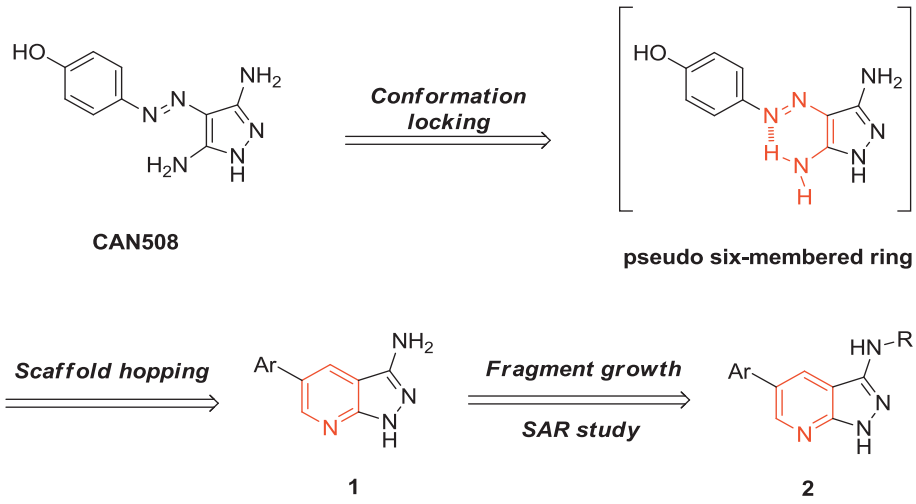

Fig. 3. Molecular design based on CAN508.

six-membered ring between the benzene and the pyrazole rings were formed via conformation locking by hydrogen bonding (Fig. 3). Accordingly, a scaffold hopping strategy was applied to generate the pyrazolo[3,4-*b*]pyridine molecule **1**, in which a pyridine ring was incorporated to replace the pseudo six-membered ring and presumably lock the molecule at the active conformation. The alteration in scaffold by eliminating the N=N moiety could also significantly increase the molecular stability. Various aromatic