Fitoterapia 92 (2014) 111–115

Contents lists available at ScienceDirect

Fitoterapia

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

Design, synthesis and biological evaluation of paclitaxel-mimics possessing only the oxetane D-ring and side chain structures

Xing-Xiu Chen [1], Feng Gao *,[1], Qi Wang, Xing Huang, Dan Wang

Department of Chinese Traditional Herbal, Agronomy College, Sichuan Agricultural University, No. 211, Huiming Road, Wenjiang Region, Chengdu 611130, PR China

ARTICLE INFO

Article history:
Received 29 August 2013
Accepted in revised form 27 October 2013
Available online 6 November 2013

Keywords:
Paclitaxel
Paclitaxel-mimics
Spirocyclic oxetane
D-ring
Side chain

ABSTRACT

Two spiro paclitaxel-mimics consisting only of an oxetane D-ring and a C-13 side chain were designed and synthesized on the basis of analysis of structure–activity relationships (SAR) of paclitaxel. In vitro microtubule-stabilizing and antiproliferative assays indicated a moderate weaker activity of the mimics than paclitaxel, but which still represented the first example of simplified paclitaxel analogues with significant anti-tumor biological activity.

1. Introduction

The natural diterpenoid paclitaxel (Taxol®), and its semi-synthetic derivatives docetaxel (Taxotere®) and cabazitaxel (Jevana®) (Fig. 1), are important chemotherapeutics in current clinical treatment of breast cancer, ovarian cancer, non-small cell lung cancer and prostate cancer [1]. They accelerate microtubule polymerization through a combination with tubulin and stabilize cell division cycle at G2/M phase, stimulating the apoptosis of tumor cells [2]. Due to their potent anti-cancer activities, medicinal chemists have conducted in-depth research on the SAR (Structure–Activity Relationship) of paclitaxel and have developed a new generation of anti-cancer taxoids with higher activity [3].

Current structural modification research on taxoids is mainly focused on changes to the taxane core or the side chain. On this basis, a large number of paclitaxel derivatives have been synthesized, and the resulting SAR have been summarized [4]. Despite the limited number of studies on the simplification of paclitaxel, some interesting results (Fig. 2) have been reported. Ojima et al. reported several paclitaxel-mimics with a simplified structure, in which the taxane core was substituted by an indolizidine scaffold, and the C-13 side chain was retained. These compounds expressed modest cytotoxic activities, but lost the activity to stabilize microtubules [5,6]. Kingston et al. designed and synthesized macrocyclic paclitaxel-mimics, which exhibited both cytotoxic and microtubule polymerization activity, based on the configuration study of T-Taxol [7,8]. Guenard et al. synthesized a series of novel compounds combining steroids with the side-chain of docetaxel, in which the taxane skeleton was replaced by a steroid scaffold. These compounds exhibited weak cytotoxic activities and showed no microtubule disassembly inhibitory activity [9]. Beau et al. synthesized a series of paclitaxel-mimics via click chemistry based on the study of the active three-dimensional conformation of paclitaxel, but without bioactivity data [10]. Although researchers have not yet prepared strongly bioactive paclitaxel-mimics, the bioactivities observed indicate their potential for SAR research of paclitaxel as well as the discovery of more active paclitaxel analogues [11–15].

Current SAR research on paclitaxel demonstrated that the unique oxetane D-ring and side chains of paclitaxel were indispensable active groups. A-seco-paclitaxel analogues synthesized by Ojima et al. showed moderate activity, although

* Corresponding author. Tel./fax: +86 28 86290870.
E-mail address: gaofeng@sicau.edu.cn (F. Gao).
[1] The two authors contributed equally to this work.

0367-326X/$ – see front matter
http://dx.doi.org/10.1016/j.fitote.2013.10.015

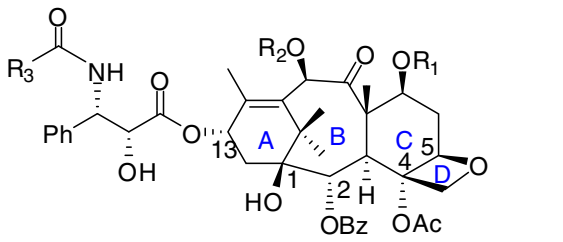

Fig. 1. Structures of paclitaxel, docetaxel and cabazitaxel.

less than paclitaxel [16]. C-seco-paclitaxel analogues, which were also obtained by Ojima et al., showed favorable activities against paclitaxel-resistant tumor cell lines [17]. In contrast, paclitaxel analogues without a D-ring or a side chain showed major reductions or even loss of activities [18–20]. In current research, two novel paclitaxel-mimics consisting only of oxetane D-rings and side chains, in which the taxane diterpenoid core was replaced by 2-oxa-6-azaspiro[3.3]heptane, were synthesized and their biological activities were evaluated.

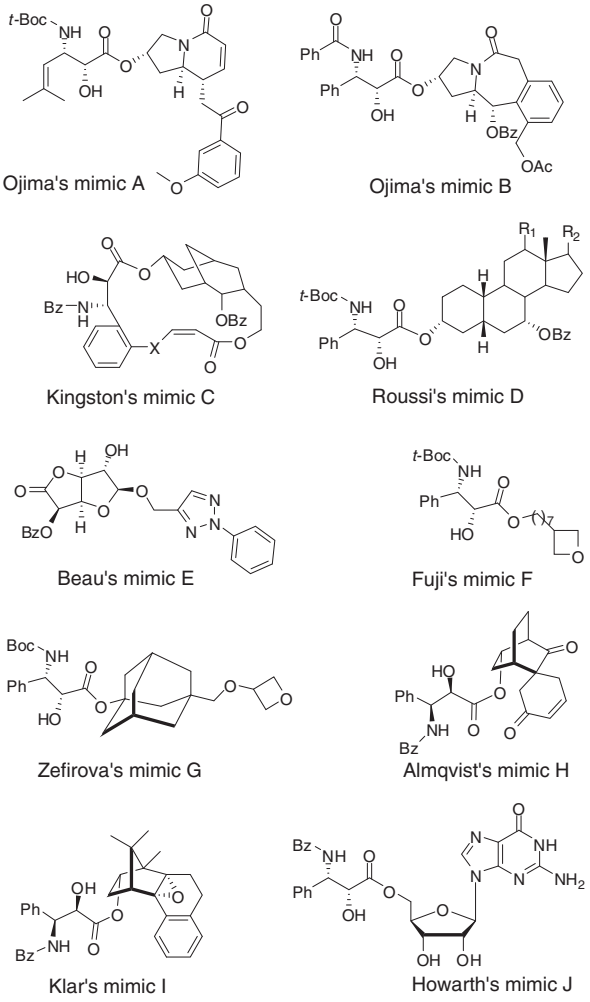

Fig. 2. Structures of representative paclitaxel-mimics.

2. Experimental

2.1. General

^{1}H and ^{13}C NMR spectra were recorded on a Varian Unity INOVA 400/54NMR spectrometer in $CDCl_3$ with tetramethylsilane (TMS) as the internal standard. Chemical shifts are given as δ value and are referenced to residual solvent proton carbon pick. Mass spectra were obtained on a VG Auto spec 3000 or on a Finnigan MAT 90 instrument. Optical rotations were measured on a Perkin-Elmer 341. Silica gel H (Qingdao Sea Chemical Factory, Qingdao, PR China) was used for column chromatography. Spots on TLC (silica gel GF_{254}) were detected with H_2SO_4–EtOH or UV. Commercially available reagents and solvents were used without further purification. Biological activity evaluation was carried out according to the protocols described previously [21].

2.2. Preparation of N-tosyl-2-oxa-6-azaspiro[3.3]heptane (5)

To a solution of KOH (179 g, 3.2 mol) and *p*-tosylamide (205 g, 1.2 mol) in 1500 mL ethanol, 3-bromo-2,2-bis (bromomethyl)propan-1-ol (324 g, 1.0 mol) was added at room temperature and the reaction mixture was heated to reflux for 90 h. The solvent was removed by evaporation, 2000 mL 1 M KOH was added and the white suspension was left to stir for another 2 h at room temperature. The mixture was filtered and the white filter cake was rinsed with water until the washing water was neutral. The filter cake was dried under high vacuum to give N-tosyl-2-oxa-6-azaspiro[3.3]heptane (5, 192 g, yield 76%) as a white solid. ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.69 (d, 2H, J = 8.4 Hz), 7.47 (d, 2H, J = 8.4 Hz), 4.42 (s, 4H), 3.85 (s, 4H), 2.41 (s, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 144.4, 130.9, 130.2, 128.5, 79.1, 59.5, 37.3, 21.3.

2.3. Preparation of 2-oxa-6-azaspiro[3.3]heptane oxalate (6)

A solution of N-tosyl-2-oxa-6-azaspiro[3.3]heptane (7.30 g, 0.0288 mol, 1.00 equiv) in MeOH (1000 mL) was sonicated, and Mg powder (5.2 g, 0.2166 mol, 9.00 equiv) was added in potions over 1 h. The crude mixture was concentrated in vacuum, and suspended in Et_2O (1000 mL) and $Na_2SO_4 \cdot 10H_2O$ (50 g) was added. The suspension was vigorously stirred at room temperature for 1 h, then filtered, the filtrate dried (Na_2SO_4), and filtered. To the filtrate was added under stirring a solution of anhydrous oxalic acid (4.5 g, 0.05 mol) in EtOH (10 mL), which immediately formed a white precipitate. The solid was filtered and dried under reduced pressure to give pure 2-oxa-6-azaspiro[3.3]heptane (3.22 g, 73%) of oxalate mono-salt as an amorphous white powder. ^{1}H NMR (400 MHz, $CDCl_3$): δ 4.65 (s, 4H), 4.12 (s, 4H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 79.2, 54.1, 39.3.