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Novel hybrids of natural β-elemene bearing isopropanolamine moieties: Synthesis, enhanced anticancer profile, and improved aqueous solubility

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ABSTRACT

A series of novel β-elemene isopropanolamine derivatives were synthesized and evaluated for their antitumor activity. The results indicated that all of the compounds showed stronger antiproliferative activities than β-elemene as well as improved aqueous solubility. In particular dimer **6q** showed the strongest cytotoxicity against four tumor cell lines (SGC-7901, HeLa, U87 and A549) with IC_{50} values ranging from 4.37 to 10.20 μM. Moreover, combination of **6q** with cisplatin exhibited a synergistic effect on these cell lines with IC_{50} values ranging from 1.21 to 2.94 μM, and reversed the resistance of A549/DPP cells with an IC_{50} value of 2.52 μM. The mechanism study revealed that **6q** caused cell cycle arrest at the G2 phase and induced apoptosis of SGC-7901 cells through a mitochondrial-dependent apoptotic pathway. Further *in vivo* study in H22 liver cancer xenograft mouse model validated the antitumor activity of **6q** with a tumor inhibitory ratio (TIR) of 60.3%, which was higher than that of β-elemene (TIR, 49.1%) at a dose of 60 mg/kg. Altogether, the potent antitumor activity of **6q** *in vitro* and *in vivo* warranted further preclinical investigation for potential anticancer chemotherapy.

1. Introduction

Over the past decade, there is a markedly increasing trend in the incidence and mortality of malignant tumors around the world [1] and cancer has become the first leading cause of death in China, beyond cerebrovascular and heart diseases [2]. The extensive prescription of synthetic antitumor drugs is being ruled out owing to their toxicity, resistance and unwanted side effects [3,4]. This led to the search for new antineoplastic agents, particularly those obtained from natural sources such as animals, plants, microbes and marine organisms [5,6]. In recent years, a large number of natural products, especially terpenes, have been discovered with marked anticancer activity *in vitro* and *in vivo*, some of which have been successfully developed for clinical use to treat human neoplastic diseases [7–9].

Curcuma wenyujin is a popular group of traditional Chinese medicine plants whose essential oils are widely used in cancer treatment in China [10]. β-Elemene (**1**, Scheme 1), a sesquiterpene compound extracted from the essential oils of *Curcuma wenyujin*, accounts for 60–72% of elemene including α, β, γ and δ forms [10]. As the major active antitumor component in the elemene mixture, β-elemene has been isolated and approved by the Chinese Food and Drug Administration for the treatment of human cancers [11]. The major advantages of β-elemene as an anticancer drug are [12–14]: i) broad-spectrum antitumor effects in various types of cancers, including drug-resistant tumors; ii) not inducing any multidrug resistance and reversing the resistance of other antitumor drugs in tumor cells; and iii) low toxicity without bone marrow suppression. Despite these striking antitumor properties, the poor water solubility and moderate activity of β-elemene hampers its wide applications in clinic.

Structural modifications are an effective approach to improve the druggability of natural compounds [15]. And it has been reported that introduction of oxygen or nitrogen-containing polar group into the skeleton of β-elemene could favorably impact its water solubility and antitumor activity [16,17]. Enlightened by these findings, we designed a series of novel β-elemene isopropanolamine derivatives by introducing both amine and hydroxyl groups to increase the water solubility and antitumor activity of natural β-elemene. Herein, we report synthesis, *in vitro* and *in vivo* antitumor activity, and anticancer mechanism for a new class of β-elemene isopropanolamine derivatives with improved aqueous solubility.

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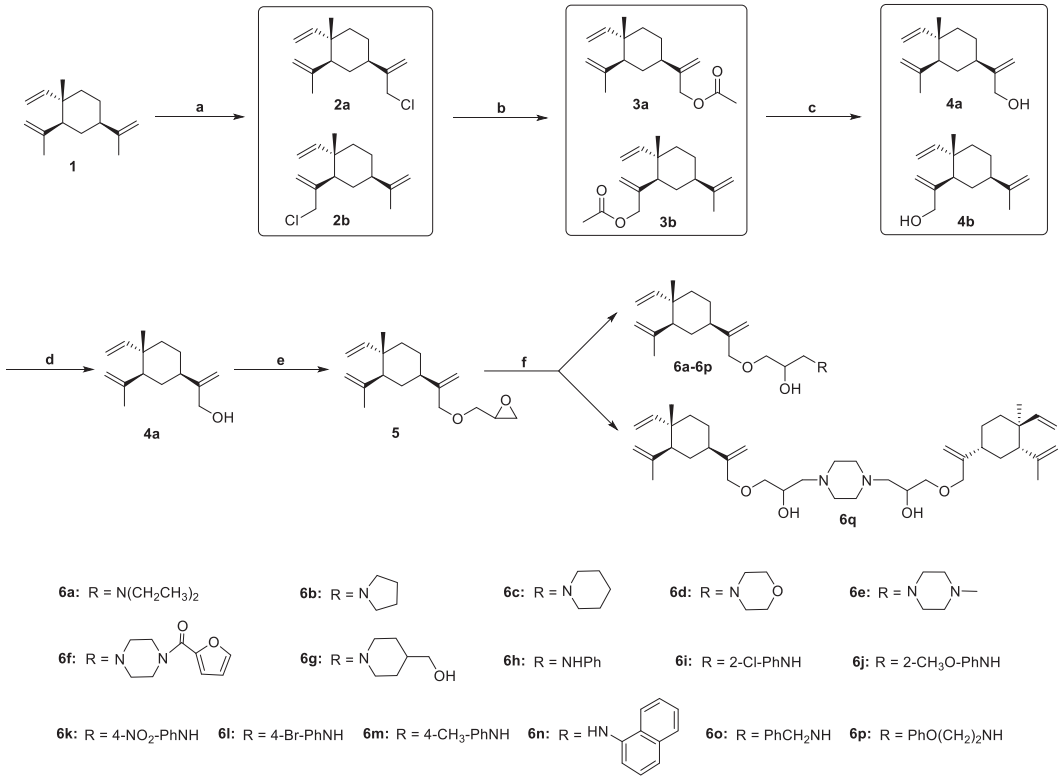

Scheme 1. Synthetic routes of the title compounds **6a–6q**. Reagents and conditions: (a) NaClO, HOAc/CH_2Cl_2, 0–5 °C, 6 h, 55%; (b) DMF, NaOAc, 120 °C, 8 h, 75%; (c) MeOH/$CHCl_3$, KOH, reflux, 2 h, 85%; (d) separated by HPLC, Hexane/EtOH = 98/2 (V/V), UV = 214 nm; (e) epibromohydrin, NaH, anhydrous DMF, rt, 3 h, 72%; (f) amines, *Cat.* $Zn(ClO_4)_2{\cdot}6H_2O$, 80 °C, 1 h, 58–84%.

2. Results and discussions

2.1. Chemistry

The synthesis of *β*-elemene isopropanolamine derivatives **6a–6q** was shown in Scheme 1. Chlorination of *β*-elemene (**1**) with NaClO produced the chlorinated mixture of **2a** and **2b**, followed by treatment with AcONa to give the acylated compounds **3a** and **3b**. The resulting products were subjected to alkaline hydrolysis to produce a mixture of 13-*β*-elemol (**4a**) and 14-*β*-elemol (**4b**), which was separated by HPLC to provide the main component **4a** [18]. Subsequent alkylation of **4a** with epibromohydrin in the presence of NaH gave the epoxide intermediate **5**, which was further reacted with different amines in the presence of a catalytic amount of $Zn(ClO_4)_2{\cdot}6H_2O$ to obtain the target compounds **6a–6q** [19].

2.2. Pharmacology

2.2.1. In vitro antiproliferative activity

Initially, *β*-elemene isopropanolamine derivatives **6a–6q** were examined for their antiproliferative activities against three cancer cell lines (SGC-7901: human gastric carcinoma; HeLa: human cervical adenocarcinoma; U87: human glioblastoma). As shown in Table 1, all of the derivatives exhibited stronger activities than parent compound *β*-elemene, and some of them even showed preferable activities than positive control cisplatin, suggesting that introduction of an isopropanolamine moiety was beneficial for the antitumor activity of *β*-elemene. Aliphatic (**6a**), naphthenic (**6b–6g**) and aryl (**6h–6n**) amines showed nearly the same activity, approximately 3- to 10-fold more potent than *β*-elemene. When the amine was benzylamine (**6o**) or phenoxyethylamine (**6p**) containing a free NH, the activity was markedly improved with IC_{50} values of 10~20 μM on all tested cell lines. It is interesting that among these derivatives, dimer **6q** exhibited the strongest activity with IC_{50} values of 4.37, 7.56 and 10.20 μM against SGC-7901, HeLa and U87 cells, respectively, which was superior to cisplatin with IC_{50} values of 9.09, 16.28 and 21.39 μM, respectively.

Table 1
Antiproliferative activities of *β*-elemene isopropanolamine derivatives against three cancer cell lines.

Compd	Cell lines (IC_{50}[a], μM)		
	SGC-7901	HeLa	U87
β-Elemene	236.27 ± 18.41	213.51 ± 15.23	179.72 ± 15.37
6a	43.32 ± 3.78	36.26 ± 3.86	49.00 ± 5.44
6b	42.86 ± 4.43	29.96 ± 1.89	35.02 ± 3.92
6c	24.33 ± 2.67	29.45 ± 3.95	37.93 ± 3.08
6d	44.87 ± 5.54	52.29 ± 4.72	39.07 ± 2.58
6e	31.25 ± 2.47	26.42 ± 0.87	23.99 ± 1.69
6f	37.75 ± 1.55	31.81 ± 2.98	38.20 ± 5.14
6g	49.23 ± 1.82	20.79 ± 3.43	22.12 ± 0.96
6h	56.76 ± 4.29	55.77 ± 6.17	56.36 ± 3.63
6i	34.42 ± 2.86	41.19 ± 3.74	38.92 ± 4.51
6j	51.89 ± 5.89	37.95 ± 3.28	66.05 ± 5.43
6k	26.41 ± 1.09	22.67 ± 1.65	28.50 ± 2.83
6l	65.16 ± 5.13	39.88 ± 4.78	54.86 ± 3.62
6m	50.27 ± 4.42	66.53 ± 3.21	61.47 ± 6.46
6n	36.49 ± 3.63	35.49 ± 1.03	57.41 ± 5.86
6o	21.40 ± 1.22	10.04 ± 0.52	12.63 ± 1.04
6p	15.40 ± 1.67	9.42 ± 0.73	10.50 ± 0.91
6q	4.37 ± 0.51	7.56 ± 0.65	10.20 ± 0.76
Cisplatin	9.09 ± 0.83	16.28 ± 1.06	21.39 ± 1.91

[a] IC_{50}: concentration of the test compound that inhibits 50% of cell growth. Results are expressed as the mean ± SD (n = 3).

2.2.2. In vitro antiproliferative activity of **6q** in combination with cisplatin

It was reported that *β*-elemene increased the sensitivity of several cancer cells and even reversed the resistance to cisplatin [20–24]. In order to investigate whether **6q** could induce sensitization to cisplatin,