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Asymmetric total synthesis and antiproliferative activity of goniothalamin oxide isomers

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ABSTRACT

Goniothalamin oxide (**1**) is a styryl lactone which was isolated from bark and leaves of several *Goniothalamus* species. This natural product has some interesting biological properties such as larvicidal and tripanocidal activities. However, no studies on the antiproliferative profile of goniothalamin oxide (**1**) and its stereoisomers have been reported yet. Here, goniothalamin epoxide (**1**), isogoniothalamin epoxide (**2**) and their enantiomers were prepared via epoxidation of (*R*)-and (*S*)-goniothalamin (**4**). A 3:2 molar ratio in favor of goniothalamin oxide (**1**) and *ent*-**1** was observed from (*R*)- and (*S*)-**4**, respectively, when 3-chloroperbenzoic acid (*m*CPBA) was employed while an increase to 6:1 molar ratio was achieved with (*S*,*S*)-Jacobsen's catalyst. Antiproliferative activity of these epoxides revealed that *ent*-isogoniothalamin oxide (*ent*-**2**) was the most active against the eight cancer cell lines studied. These results indicate that 6*S*, 7*R* and 8*R* absolute configurations are beneficial for the activity of these epoxides.

1. Introduction

Phytochemical studies of the genus *Goniothalamus* have resulted in the isolation and characterization of many compounds with a variety of biological activity [1–4]. Goniothalamin oxide (**1**) (Fig. 1) is a styryl lactone that was isolated from the bark and leaves of *Goniothalamus macrophyllus* [5], *Goniothalamus amuyon* [6], and *Goniothalamus dolichocarpus* [7]. The absolute configuration of this natural product was determined as being (6*R*, 7*R*, 8*R*) after correlation with data from X-ray diffraction studies of the synthetic derivative isogoniothalamin oxide (**2**) (Fig. 1) [7]. Goniothalamin oxide (**1**) has demonstrated to display high toxicity against larvae of *Aedes aegypti* requiring low concentration to kill 50% of the larvae (LC_{50} = 50–100 μg/mL). Recently, we have described the tripanocidal activity of goniothalamin oxide (**1**) and isogoniothalamin oxide (**2**) [8]. Our results have shown that both epoxides (IC_{50} = 0.25 mM) have similar potency to promote lyses of the trypomastigote forms of *Trypanosoma cruzi* [8]. In addition to its larvicidal and tripanocidal activities, goniothalamin oxide (**1**) was identified to have embryotoxic and teratogenic activities as reported by Sam and coworkers [5].

These properties associated to goniothalamin oxide (**1**) must be taken cautiously because it is well known that antiproliferative compounds may present significant embryotoxic and teratogenic activities [9,10]. In fact, no study on the biological profile, particularly those related to the antiproliferative activity of goniothalamin oxide (**1**) as well as its stereoisomers has been reported yet.

The synthetic efforts towards goniothalamin oxide (**1**) and isogoniothalamin oxide (**2**) reported so far are generally carried out through epoxidation of (*R*)-goniothalamin [(*R*)-(**4**)] available from natural or synthetic origin [5,7,11–15].

Herein, we report the synthesis of goniothalamin oxide (**1**), isogoniothalamin oxide (**2**) and their respective enantiomers (*ent*-**1** and *ent*-**2**) featuring epoxidation reaction using 3-chloroperbenzoic acid (*m*CPBA) or (*R*,*R*)- or (*S*,*S*)-*N*,*N*′-bis(3,5-di-*tert*-butylsalicylidene)-1,2-cyclohexane diaminomanganese(III) chloride (Jacobsen's catalyst) (Scheme 1). Additionally, the antiproliferative activities of goniothalamin oxide (**1**), isogoniothalamin oxide (**2**) and their respective enantiomers (*ent*-**1** and *ent*-**2**) against cancer cell lines are also described.

2. Materials and methods

2.1. Chemistry

2.1.1. General procedures

Reagents and solvents are commercial grade and were used as supplied, except dichloromethane and triethylamine which were distilled from calcium hydride. Chromatographic separations were

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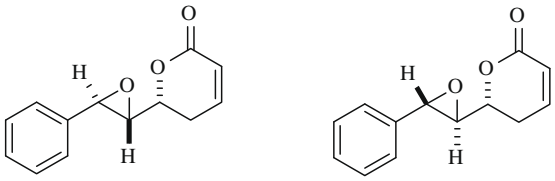


Fig. 1. Structures of goniothalamin oxide (**1**) and isogoniothalamin oxide (**2**).

performed using 70–230 Mesh silica gel. Thin-layer chromatography was carried out on Macherey–Nagel precoated silica plates (0.25 mm layer thickness). IR spectra were obtained on Nicolet Impact 410 FT (film or KBr). ^{1}H NMR and ^{13}C NMR data were recorded on a Varian Gemini 2000 (7.0 T) or Varian Inova 500 (11.7 T) spectrometer. Chemical shifts are reported in δ (ppm) relative to $(CH_3)_4Si$ for ^{1}H-NMR and to $CDCl_3$ for ^{13}C-NMR. For ^{1}H NMR, the chemical shifts were followed by multiplicity (s, singlet; d, doublet; dd, double dublet; ddd, double double dublet; t, triplet; q, quartet; m, multiplet) and coupling constant *J* reported in Hertz (Hz). High-resolution mass spectra (HRMS) were measured on a VG Autospec-Micromass spectrometer. Chiral GC analyses were performed with capillary column CP-Chirasil-DEX CB fused silica WCOT (25 m × 0.25 mm × 0.25 μm) on Agilent 6890 series GC system. Optical rotations were measured at 25 °C with Perkin–Elmer 241 instrument.

*2.1.2. Preparation of goniothalamin oxide (**1**) and its isomers (**2**, ent-**1** and ent-**2**) using mCPBA as oxide agent*

To a solution of 95.6 mg (0.48 mmol) of (*R*)-goniothalamin [(*R*)-(**4**)] in 14.3 mL of CH_2Cl_2 was added *m*CPBA (70% W/W, 141.2 mg, 0.57 mmol) and the mixture was stirred at room temperature. After 5 h, saturated $NaHCO_3$ was added until no evolution of gas was observed. The solution was washed with ether (3 × 20 mL), the organic phases were separated and dried with anhydrous $MgSO_4$. After solvent removal, the residue was purified by column chromatography on silica gel using hexane/ethyl acetate (3:2, v/v) as eluent. **1** and **2** were obtained in 64% yield (3:2 molar ratio, respectively). The same procedure using (*S*)-goniothalamin [(*S*)-(**4**)] afforded *ent*-**1** and *ent*-**2** (same ratio and yield). Goniothalamin oxide (**1**) and its isomers (**2**, *ent*-**1** and *ent*-**2**) where obtained as white solids. Goniothalamin oxide (**1**) and *ent*-**1**: mp = 90.1–93.8 °C. IR (KBr): 3420, 3062, 2960, 1724, 1631, 1466, 1398, 1377, 1259, 1043, 812, 704 cm^{-1}. ^{1}H-NMR (300 MHz, $CDCl_3$): δ 7.39 – 7.25 (m, 5H), 6.94 (ddd, 1H, *J* 9.6, 4.6 and 3.8 Hz), 6.07 (dt, 1H, *J* 9.9 and 1.8 Hz), 4.44 (dt, 1H, *J* 9.6 and 5.6 Hz), 3.89 (d, 1H, *J* 1.8 Hz), 3.27 (dd, 1H, *J* 5.4 and 1.8 Hz), 2.62 – 2.56 (m, 2H). ^{13}C-MNR (125 MHz, $CDCl_3$): δ 162.8, 144.3, 135.6, 128.7, 128.6 (2C), 125.7 (2C), 121.5, 77.2, 61.5, 57.2, 25.9. HRMS (EI) *m/z* calculated M^+ = 216.07864, found 216.07802. $[\alpha]_D^{25}1 = +113.0$ (*c* 0.8; $CHCl_3$), $[\alpha]_D^{25}$ of *ent*-**1** = −121.0 (*c* 0.7, $CHCl_3$). Isogoniothalamin oxide (**2**) and *ent*-**2**: mp = 112.0–113.4 °C. IR (KBr): 3072, 2962, 1726, 1383, 1254, 1074, 1032, 879, 818 cm^{-1}. ^{1}H-NMR (300 MHz, $CDCl_3$): δ 7.41 – 7.27 (m, 5H), 6.94 (ddd, 1H, *J* 9.9, 5.5 and 3.1 Hz), 6.08 (ddd, 1H, *J* 9.9, 1.1 and 0.9 Hz), 4.70 (dt, 1H, *J* 10.5 and 4.3 Hz), 4.10 (d, 1H, *J* 2.4 Hz), 3.25 (dd, 1H, *J* 3.4 and 1.9 Hz), 2.74–2.50 (m, 2H). ^{13}C-MNR (125 MHz, $CDCl_3$): δ 162.9, 144.0, 135.8, 128.6 (3C), 125.6 (2C), 121.5, 75.1, 62.1, 55.0, 26.1. HRMS (EI) *m/z* calculated M^+ = 216.07864, found 216.07344. $[\alpha]_D^{25}$ of **2** = +31.3 (*c* 1.35, $CHCl_3$), $[\alpha]_D^{25}$ of *ent*-**2** = −32.0 (*c* 0.9, $CHCl_3$).

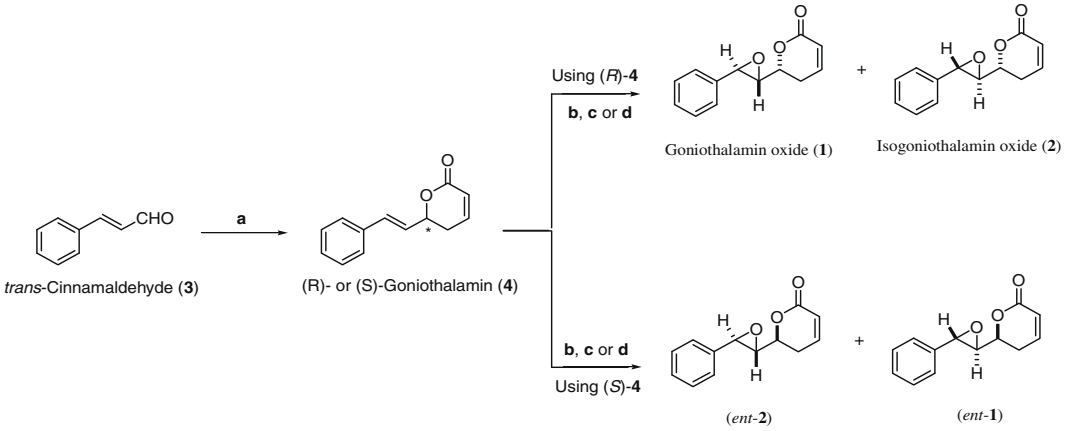
*2.1.3. Preparation of goniothalamin oxide (**1**) and its isomers (**2**, ent-**1** and ent-**2**) using Jacobsen's catalyst*

A solution of 0.05 mol.L^{-1} of Na_2HPO_4 (25 mL) was added to 5 mL of commercially available household bleach. The pH of the resulting buffered solution was adjusted to pH 11.3 by addition of a few drops of 1 mol.L^{-1} NaOH or HCl depending on the commercial solution of NaOCl used. This solution was cooled to 0 °C and then added at once to a 0 °C solution containing 50 mg (0.25 mmol) of (*R*)-goniothalamin [(*R*)-(**4**)] and 15.5 mg (0.025 mmol) of (*R,R*)-*N,N′*-bis(3,5-di-*tert*-butylsalicylidene)-1,2-cyclohexane diaminomanganese(III) chloride in 0.25 mL of CH_2Cl_2. The two-phase mixture was stirred at room temperature, and the reaction progress was monitored by TLC. After 5 h, 20 mL of saturated $NaHCO_3$ was added and this mixture was washed once with 20 mL of ether. The organic phase was separated and aqueous phase was washed twice with ether (3 × 20 mL). The organic phases were dried with anhydrous $MgSO_4$. After solvent removal, the residue was purified by chromatography on silica gel using hexane/ethyl acetate (3:2, v/v) as eluent. The products **1** and **2** were obtained in 52% yield and 2.3:1 molar ratio, respectively. The same procedure using (*S,S*)-*N,N′*-bis(3,5-di-*tert*-butylsalicylidene)-1,2-cyclohexane diaminomanganese(III) chloride furnished **1** and **2** in 49% yield and 6:1 molar ratio.

2.2. Biological activities

2.2.1. Biological assay

Human tumor cell lines UACC-62 (melanoma), MCF-7 (breast), NCI-H460 (lung, non-small cells), OVCAR-03 (ovarian), PC-3 (prostate), HT-29 (colon), 786-0 (renal) and NCI-ADR/RES (ovarian



Scheme 1. Synthesis of goniothalamin oxide (**1**), *ent*-**1**, isogoniothalamin oxide (**2**), and *ent*-**2** from (*R*)- or (*S*)-goniothalamin (**4**). *Conditions*: (a) see Ref. [16–19]; (b) *m*CPBA (70%), CH_2Cl_2, room temperature, 5 h (61–64%); (c) (*R,R*)-Jacobsen, CH_2Cl_2; Na_2HPO_4 (0.05 mol/L) and commercial household bleach, pH 11.3 at 0 °C, then room temperature, 5 h (49–52%); (d) (*S,S*)-Jacobsen, CH_2Cl_2; Na_2HPO_4 (0.05 mol/L) and commercial household bleach, pH 11.3 at 0 °C, then room temperature, 5 h (49–52%).

expressing phenotype multiple drugs resistance) were kindly provided by Frederick Cancer Research & Development Center – National Cancer Institute – Frederick, MA, USA. Stock cultures were grown in 5 mL of RPMI 1640 (GIBCO BRL, Life Technologies) supplemented with 5% of fetal bovine serum. Gentamicine (50 μg/mL) was added to the experimental cultures. Cells in 96-well plates (100 μL cells/well) were exposed to various concentrations of epoxides **1**, *ent*-**1**, **2** and *ent*-**2** DMSO (0.25, 2.5, 25 and 250 μg/mL) at 37 °C, 5% of CO_2 for 48 h. The final concentration of DMSO did not affect the cell viability. Then, a 50% of trichloroacetic acid solution was added and after incubation (30 min at 4 °C), washing and drying, the cell proliferation was determined by spectrophotometric quantification (540 nm) of cellular protein content using sulforhodamine B assay [16]. The background absorbencies were subtracted from the appropriate control and drug-blank measurements. To assess the effect of goniothalamin oxide (**1**) and its isomers (**2**, *ent*-**1** and *ent*-**2**) on cell growth, three measurements were obtained at time zero (T_0) values for all cells at the beginning of incubation, and control (C) and test (T) values at the end of incubation without and with the test substance, respectively. For T value $\leqslant T_0$ (cytostatic effect), the calculation was $100x\,[(T - T_0)/C - T_0]$. While for $T < T_0$ (cytocidal effect), the calculation was $100x[(T - T_0)/T_0]$. The GI_{50} values (test substance concentration eliciting 50% inhibition of cell growth) and TGI (test substance concentration eliciting 100% inhibition of cell growth) were determined by non-linear regression analysis using Origin software, version 7.5. These results presented here refer to a representative experiment since all assays were run in triplicate and the average standard error was always <5%.

3. Results and discussion

3.1. Chemistry

Goniothalamin oxide (**1**), isogoniothalamin oxide (**2**) and their respective enantiomers (*ent*-**1** and *ent*-**2**) were obtained from the epoxidation of goniothalamin enantiomers, prepared in three steps from *trans*-cinnamaldehyde, as previously described (Scheme 1) [17–20].

Epoxidation of enantiomerically enriched (>95% enantiomeric excess by chiral gas chromatography) (*R*)-goniothalamin (**4**) with commercially available 70% *m*-chloroperbenzoic acid (*m*CPBA) under standard conditions (1.2 equiv., CH_2Cl_2, rt) provided a 3:2 molar ratio of goniothalamin oxide (**1**) and isogoniothalamin oxide (**2**) in 64% yield, after separation by column chromatography on silica gel. The *trans* configuration of epoxides **1** and **2** was assigned by the inspection of the coupling constant between H_7 and H_8 (1.8 and 2.4 Hz, respectively). The stereochemistry of each diastereoisomer was established by comparison with literature data [5]. Despite the good correlation of the NMR data of the two diastereoisomers formed in the *m*CPBA epoxidation of (*R*)-goniothalamin (**4**) with those described by Sam and coworkers [5], the optical rotation of isogoniothalamin oxide (**2**) prepared by us {$[\alpha]_D$ = +31.3 (*c* 1.3, $CHCl_3$)} was significantly different from that described in the above reference {$[\alpha]_D$ = −106.0 (*c* 1.5, $CHCl_3$)}. Considering that the specific optical rotation found by us for goniothalamin oxide (**1**) {$[\alpha]_D^{25} = +113.0$ (*c* 0.8; $CHCl_3$)} nicely matches the one described in the literature by Sam and coworkers {($[\alpha]_D^{25} = +107.0$ (*c* 0.7; $CHCl_3$)}, we consider that a revision of the specific optical rotation for isogoniothalamin oxide (**2**) may be in order. The stereoselective epoxidation of (*R*)-goniothalamin (**4**) is consistent with the reaction taking place at the *Si* face of the styrenic double bond to afford the major goniothalamin epoxide (**1**).

Accordingly, when enantiomerically enriched (>95% enantiomeric excess) (*S*)-goniothalamin (**4**) was employed under the same epoxidation conditions, a 3:2 molar mixture of *ent*-goniothalamin oxide (*ent*-**1**) and *ent*-isogoniothalamin oxide (*ent*-**2**) was formed {(*ent*-**1**: $[\alpha]_D^{25} = -121.0$ (*c* 0.7, $CHCl_3$); *ent*-**2**: $[\alpha]_D^{25} = -32.0$ (*c* 0.9, $CHCl_3$)}, respectively). It is to be noticed that the absolute value of the specific optical rotation for *ent*-**2** again differs from that described in the literature for isogoniothalamin oxide (**2**) but is in full agreement with the one observed in this work.

Markó and coworkers have shown that (*R*)-(+)-goniothalamin oxide (**1**) was prepared in 19:1 diastereoisomeric ratio and 98% yield when purified and water free *m*CPBA (4.0 equiv.) was employed in CH_2Cl_2 at 0 °C [14]. Similar results were recently described by Bose and coworkers [15]. However, both Markó and Bose also reported that the use of commercially available 70% *m*CPBA in CH_2Cl_2 at reflux provided a 3:2 mixture of (*R*)-goniothalamin oxide (**1**) and (*R*)-isogoniothalamin oxide (**2**), in 69% yield. In order to secure both enantiomeric forms of goniothalamin oxide (**1**) and isogoniothalamin oxide (**2**) for biological evaluation, we have initially employed commercially available 70% w/w *m*CPBA (1.2 equiv.) in CH_2Cl_2 at rt which in our hands provided a 3:2 mixture of the two diastereoisomers, in 64% overall yield. The diastereoisomeric ratio could be determined by inspection of the ^{1}H-NMR spectrum of the crude mixture and it was confirmed after isolation of goniothalamin oxide (**1**) and isogoniothalamin oxide (**2**) by chromatographic separation on silica gel.

We have investigated the use of Jacobsen's catalyst in order to introduce chiral discrimination by the reagent [21] aiming to prepare the enantiomers of isogoniothalamin oxide as the major epoxides. However, when (*R,R*)-Jacobsen's catalyst was employed in the epoxidation of (*R*)-**4** only a slight increase in the diastereoisomeric ratio in favor of goniothalamin oxide (**1**) was observed (*d.r.* = 2.3:1, 52% yield). The use of (*S,S*)-Jacobsen's catalyst afforded a 6:1 molar ratio of goniothalamin oxide (**1**) and isogoniothalamin oxide (**2**) which were obtained in a combined 49% yield. Bose and coworkers [15] have also employed (S,S)-Jacobsen's catalyst in the epoxidation of (*R*)-goniothalamin (**4**) but under different experimental conditions compared to those described here: either hexafluoroacetone or acetone were employed in the presence of Oxone® in tetrabutylammonium sulfate buffered solution and acetonitrile as solvent to afford (*R*)-goniothalamin oxide as the major product (98:2 and 95:5 ratio, respectively) in very good yields (90% and 80% yield).

Considering the difference in the experimental conditions employed by us and those described by Bose and coworkers [15], it is not easy to rationalize the differences observed in both yield and diastereoisomeric ratios. In any event, these results indicate that the use of Jacobsen's catalysts in the epoxidation of goniothalamin is not an efficient route to isogoniothalamin epoxide (**2**) and are in accordance with those described by Katsuki and coworkers for the epoxidation of *trans* double bonds by optically active (salen) manganese (III) complexes derived from (*S,S*)-1,2-diaminocyclohexane where preferencial epoxidation of the *Si* face of the *trans* styrenic double bond (regarding the ipso carbon) was observed albeit in low *e.e.* [22]. In our case, it seems that chiral discrimination of the diastereotopic faces of the styrenic double bond is enhanced by the intrinsic *Si* face preference of the substrate (matched pair).

3.2. Biological activities

Since it is well known that different cell lines display different sensitivities toward a cytotoxic compound, in the present study we have used cell lines of various histological origin [MCF-7 (breast), NCI-ADR/RES (ovarian expressing the resistance phenotype for adryamycin), NCI-H460 (lung, non-small cells), UACC-62 (melanoma), 786-0 (kidney), OVCAR-03 (ovarian), PC-3 (prostate), and HT-29 (colon)] for the initial evaluation of the cytotoxicity of goniothalamin oxide (**1**), isogoniothalamin oxide (**2**) and its respective enantiomers. Cell proliferation was determined by spectrophotometric assay using sulforhodamine B as protein-binding

dye. Epoxides **1**, **2**, *ent*-**1**, and *ent*-**2** were used at 0.25–250 μg/mL and doxorubicin (DOX) at the same concentration range as positive control (see Experimental Section for details). Concentration that elicits 50% and 100% inhibition of cell growth (GI_{50} and TGI, respectively) were determined after 48 h of cell treatment. As shown in Fig. 2, goniothalamin oxide (**1**), isogoniothalamin oxide (**2**) and its respective enantiomers displayed antiproliferative activity in a concentration-dependent way against the cancer cell lines tested. Epoxides **1**, **2**, and *ent*-**2** at 2.5 μg/mL had only a cytostatic activity against all cancer cell lines studied (Fig. 2). On the other hand, *ent*-**1** at the same concentration presented cytotoxic activity against UACC-62 (melanoma), OVCAR-03 (ovarian), HT-29 (colon), and 786-0 (kidney) cancer cell lines. At concentrations higher than 2.5 μg/mL all epoxides were cytotoxic for all cancer cell lines evaluated (Fig. 2). GI_{50} and TGI values (μg/mL) for **1**, **2**, *ent*-**1**, and *ent*-**2** as well as DOX are summarized in Table 1.

It is noteworthy that among the four epoxides evaluated, *ent*-**2** was the most potent against the cancer cell lines studied (Table 1). These results indicate that the 6*S* absolute configuration is an important structural requirement to antiproliferative activity of *ent*-**2**. In fact, *ent*-**2** was 7- and 8-fold more potent than **1** and *ent*-**1** to inhibit the total growth of melanoma (UACC.62) cancer cells, respectively. Additionally, *ent*-**2** was 5-fold more potent than **2** in inhibiting the total growth of melanome (UACC.62) cancer cells. Additionally, the total growth inhibition (TGI) for kidney cancer cells (786-0) in the presence of *ent*-**2** was significantly lower (4- to 5-fold) than when it was exposed to the other diastereoisomers. In breast cancer cells (MCF-7), the TGI values for **1**, *ent*-**1** and **2** were 2- to 4-fold higher than the corresponding value determined for *ent*-**2**. Ovarian expressing the resistance phenotype for adryamycin (NCI-ADR/RES), lung, non-small cells (NCI-H460) and ovarian (OVCAR-03) cancer cells were more sensitive when treated with *ent*-**2** than **1**,

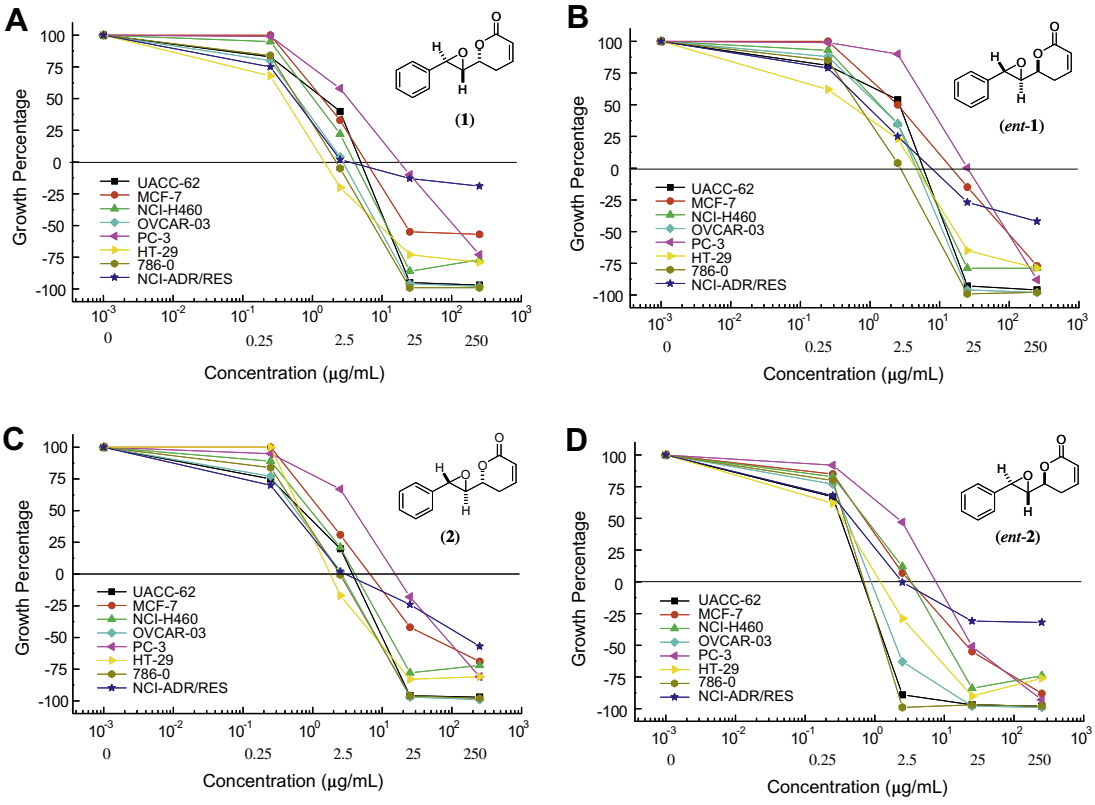


Fig. 2. Percentage growth of cancer cell for 48 h with different concentrations (0.25, 2.5, 25 and 250 μg/mL) of epoxides **1**, *ent*-**1**, **2**, and *ent*-**2** against cancer cell lines. (A) goniothalamin oxide (**1**), (B) epoxide *ent*-**1**, (C) isogoniothalamin oxide (**2**), and (D) epoxide *ent*-**2**. The experiments were done in triplicate as described in experimental section using a panel of NCI human tumoral cells. MCF-7 (breast), NCI-ADR/RES (ovarian expressing the resistance phenotype for adryamycin), NCI-H460 (lung, non-small cells), UACC-62 (melanoma), 786-0 (kidney), OVCAR-03 (ovarian), PC-3 (prostate), and HT-29 (colon).

Table 1
Comparison of antiproliferative activity, in μg/mL, of epoxides **1**, *ent*-**1**, **2**, *ent*-**2** against cancer cell lines.

Compound	UACC-62		MCF-7		NCI-H460		OVCAR-03		PC-3		HT-29		786-0		NCI-ADR/RES	
	GI_{50}[a]	TGI[b]	GI_{50}[a]	TGI[b]	GI_{50}[a]	TGI[b]	GI_{50}[a]	TGI[b]	GI_{50}[a]	TGI[b]	GI_{50}[a]	TGI[b]	GI_{50}[a]	TGI[b]	GI_{50}[a]	TGI[b]
1	2.46	4.02	2.25	7.07	1.93	3.87	0.50	2.51	2.55	18.47	0.27	1.74	0.32	2.24	0.41	17.73
ent-1	2.50	4.75	2.50	15.06	2.26	4.59	2.40	3.74	4.52	25.22	0.41	3.76	0.56	2.56	0.82	11.83
2	0.58	3.16	2.21	7.92	1.04	4.03	0.43	2.38	2.76	16.53	0.63	2.06	0.35	2.39	0.37	6.28
ent-2	0.27	0.57	0.60	3.76	0.64	3.30	0.29	0.94	2.49	7.46	0.26	1.35	0.29	0.55	0.29	7.56
DOX[c]	0.02	0.19	0.20	2.68	0.02	0.11	0.19	12.91	0.14	0.41	0.25	2.26	0.03	0.25	0.09	5.31

[a] Concentrations that elicit inhibition by 50% of the cell growth (GI_{50} in μg/mL).
[b] Concentrations that elicit inhibition by 100% of the cell growth (TGI in μg/mL) were determined from non-linear regression analysis using the Origin 7.5 software.
[c] Doxorubicin (DOX) was employed as positive control.

ent-**1** and **2**. Prostate (PC-3) cancer cells were inhibited in same extension when treated with **1**, *ent*-**2** and **2**. Goniothalamin oxide (**1**) and *ent*-**2** were as potent as DOX in inhibiting the growth of colon (HT-29) cancer cells (Table 1). In addition to this, the GI_{50} value for *ent*-**2** was roughly the same of that for DOX against ovarian (OVCAR-03) cancer cells while total growth inhibition occurred at significant lower concentration (0.94 μg/mL) when *ent*-**2** was employed as compared to doxorubicin (12.9 μg/mL). It is noteworthy that the most sensible lineages (786-0, UACC-62, HT29) are not hormone dependent, an information that might prove to be useful in the studies on the mechanism of action of these compounds.

4. Conclusion

Goniothalamin oxide (**1**), isogoniothalamin oxide (**2**) and their enantiomers were prepared via epoxidation of (*R*)-and (*S*)-goniothalamin (**4**) with *m*CPBA A 3:2 molar ratio in favor of goniothalamin oxide (**1**) and *ent*-**1** was observed from (*R*)- and (*S*)-**4**, respectively, while an increase to 6:1 molar ratio was achieved when (*S*,*S*)-Jacobsen's catalyst was employed. Among all epoxides, *ent*-**2** was the most active against the eight cancer cell lines studied. These results indicate that 6*S*, 7*R* and 8*R* absolute configurations are beneficial for the antiproliferative activity of these epoxides.

Acknowledgments

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References

[1] M.A. Blázquez, A. Bermejo, M.C. Zafra-Polo, D. Cortes, Phytochem. Anal. 10 (1999) 161–170.
[2] Â. de Fátima, L.V. Modolo, L.S. Conegero, R.A. Pilli, C.V. Ferreira, L.K. Kohn, J.E. de Carvalho, Curr. Med. Chem. 13 (2006) 3371–3384.
[3] Â. de Fátima, W.F. Zambuzzi, L.V. Modolo, C.A.B. Tarsitano, F.R. Gadelha, S. Hyslop, J.E. de Carvalho, I. Salgado, C.V. Ferreira, R.A. Pilli, Chem. Biol. Int. 176 (2008) 143–150.
[4] C.V.B. Martins, M.A. de Resende, T.F.F. Magalhães, B.H.S. Lima, G.A. Watanabe, A.L.T.G. Ruiz, J.E. de Carvalho, R.A. Pilli, Â. de Fátima, Lett. Drug. Des. Discov. 5 (2008) 74–78.
[5] T.W. Sam, C. Sew-Yeu, S. Matsjeh, E.K. Gan, D. Razak, A.L. Mohamed, Tetrahedron Lett. 28 (1987) 2541–2544.
[6] Y.H. Lan, F.R. Chang, J.H. Yu, Y.L. Yang, Y.L. Chang, S.J. Lee, Y.C. Wu, J. Nat. Prod. 66 (2003) 487–490.
[7] S.H. Goh, G.C.L. Ee, C.H. Chuah, C. Wei, Aust. J. Chem. 48 (1995) 199–205.
[8] Â. de Fátima, C. Marquissolo, S. de Albuquerque, A.A. Carraro-Abrahão, R.A. Pilli, Eur. J. Med. Chem. 41 (2006) 1210–1213.
[9] M.V. Blagosklonny, Cell Cycle 4 (2005) 1518–1521.
[10] B. Weisz, D. Meirow, E. Schiff, M. Lishner, Expert Rev. Anticancer Ther. 4 (2004) 889–902.
[11] C. Fuganti, G. Pedrocchi-Fantoni, S. Sarra, S. Servi, Tetrahedron: Asymmetry 5 (1994) 1135–1138.
[12] J.-P. Surivet, J.-M. Vatèle, Tetrahedron 55 (1999) 13011–13028.
[13] X. Peng, A. Li, H. Shen, T. Wu, X. Pan, J. Chem. Res.(S) (2002) 330–332.
[14] J. Pospíšil, I.E. Markó, Tetrahedron Lett. 47 (2006) 5933–5937.
[15] D.S. Bose, A.V.N. Reddy, B. Srikanth, Synthesis (2008) 2323–2326.
[16] A. Monks, D. Scudeiro, P. Skehan, R. Shoemaker, K. Paull, D. Vistica, C. Hose, J. Langley, P. Cronise, A. Vaigro-Wolff, M. Gray-Goodrich, H. Campbell, J. Mayo, M. Boyd, J. Nat. Can. Inst. 83 (1991) 757–766.
[17] Â. de Fátima, R.A. Pilli, Tetrahedron Lett. 44 (2003) 8721–8724.
[18] Â. de Fátima, R.A. Pilli, Arkivoc 10 (2003) 118–126.
[19] Â. de Fátima, L.K. Kohn, M.A. Antônio, J.E. de Carvalho, R.A. Pilli, Bioorg. Med. Chem. 12 (2004) 5437–5442.
[20] Â. de Fátima, L.K. Kohn, J.E. de Carvalho, R.A. Pilli, Bioorg. Med. Chem. 14 (2006) 622–631.
[21] E.N. Jacobsen, W. Zhang, A.R. Muci, J.R. Ecker, L. Deng, J. Am. Chem. Soc. 113 (1991) 7063–7064.
[22] N. Hosoya, R. Irie, T. Katsuki, Synlett (1993) 261–263.