SUPPLEMENTARY MATERIAL

Cytotoxicity of an Unprecedented Brominated Oleanolide and a New Furoceramide from the Cameroonian Spice, *Echinops giganteus*.

Louis P. Sandjo^{a,*}, Victor Kuete^{b,c}, Xavier S. Noundou^d, Herve M. P. Poumale^a, and Thomas Efferth^{c*}

^aDepartment of Pharmaceutical Sciences, CCS –Universidade Federal de Santa Catarina, Trindade, 88040-900 - Florianopolis, SC, Brazil.

^bDepartment of Biochemistry, University of Dschang, P.O. Box 67, Dschang, Cameroon

^cDepartment of Pharmaceutical Biology, Institute of Pharmacy and Biochemistry, University of Mainz, Staudinger Weg 5, 55128 Mainz, Germany

^dDepartment of Chemical Technology, University of Johannesburg, P.O. Box 17011, Doorfontein, Johannesburg 2028, South Africa

* Authors for correspondence

Dr. Louis P. Sandjo E-mail: p.l.sandjo@ufsc.br

Prof. Thomas Efferth Phone: +49-6131-3925751; Fax: +49-6131-39-23752; E-mail: efferth@uni-mainz.de

Abstract

A preliminary study on *Echinops giganteus* (Asteraceae) showed that the methanolic extract has interesting cytotoxicities against a panel of cancer cell lines. From this extract, a lignan, a flavonoid and a polyacetylenic thiophene identified were 3 folds less cytotoxic than the extract. In the search of the metabolites responsible for the bioactivity, a new harvested *E. giganteus* was subjected to a phytochemical study using chromatographic methods. In the course of the work, two new compounds: a brominated oleanolide (**1**) and a tetrahydrofurano-ceramide (**2**) were obtained along with β -amyrin acetate (**3**), 2-(penta-1,3-diynyl)-5-(4-hydroxybut-1-ynyl)-thiophene (**4**), 2-(penta-1,3-diynyl)-5-(3,4-dihydroxybut-1-ynyl)-thiophene (**5**) and 4-hydroxy-2,6-di-(3',4'-dimethoxyphenyl)-3,7-dioxabicyclo-(3.3.0)octane (**6**). Their structures were determined on the basis of NMR spectroscopy and mass spectrometry data in conjunction with those reported in the literature. The cytotoxicity of **1**, **2**, and **5** was evaluated by employing resazurin assay against a panel of cancer cell lines with IC₅₀ values in range 6.12±0.46 - 46.96±3.61 μ M.

Keywords: Asteraceae; *Echinops giganteus*; brominated triterpene; furoceramide; cytotoxicity

Figure S1 Mass spectrum of compound **1**

Figure S2 ^1H NMR spectrum of compound **1**

Figure S3 ^{13}C NMR spectrum of compound **1**

Figure S4 COSY spectrum of compound **1**

Figure S5 HSQC spectrum of compound **1**

Figure S6 HMBC spectrum of compound **1**

Figure S7 NOESY spectrum of compound **1**

Figure S8 ¹H NMR spectrum of compound **2**

Figure S9 ¹³C NMR spectrum of compound **2**

Figure S10 COSY spectrum of compound **2**

Figure S11 HSQC spectrum of compound **2**

Figure S12 HMBC spectrum of compound **2**

Figure S13 NOESY spectrum of compound **2**

Figure S14 COSY, HMBC and NOESY correlations of compound **1**

Figure S15 COSY, HMBC and NOESY correlations of compound **2**

Figure S16 Position of the double bond in compound **2**

Figure S17 Proposal of the biosynthetic pathway of compound **1** (Thimmappa et al., 2014)

Figure S18 ESI Tandem MS spectrum of compound **2**, m/z 678.6 $[M+H]^+$

Figure S19 Fragmentation pattern of compound **2** (part 1)

Figure S20 Fragmentation pattern of compound **2** (part 2)

Reference

Thimmappa R, Geisler K, Louveau T, O'Maille P, Osbourn A. 2014. Triterpene Biosynthesis in Plants. *Annu. Rev. Plant Biol.* 65:225-257