2009

Structural revision of stemoburkilline from an E-Alkene to a Z-Alkene

Kwankamol Sastraruji
University of Wollongong

Stephen G. Pyne
University of Wollongong, spyne@uow.edu.au

Alison T. Ung
University of Wollongong, alison_ung@uow.edu.au

Pitchaya Mungkornawawakul
University of Wollongong

Wilford Lie
University of Wollongong, wilford@uow.edu.au

See next page for additional authors

Publication Details
Structural revision of stemoburkilline from an E-Alkene to a Z-Alkene

Abstract
Semisynthesis studies starting from (11Z)-1',2'-didehydrostemofoline indicated that the known Stemona alkaloid stemoburkilline is the Z-isomer and not the E-isomer as initially reported. The semisynthesis involved conversion of (11Z)-1',2'-didehydrostemofoline to II(S), 12(S)-dihydrostemofoline followed by a stereoselective base-catalyzed ring-opening reaction to give (Z)-stemoburkilline. The same product was obtained using a similar synthetic protocol starting from isostemofoline via a based-catalyzed ring-opening reaction of II (S), 12(R)-dihydrostemofoline. A re-examination of the crude root extracts of Stemona burkillii Prain and further NOE studies established stemoburkilline as the Z-isomer.

Keywords
CMMB

Disciplines
Life Sciences | Physical Sciences and Mathematics | Social and Behavioral Sciences

Publication Details

Authors
Kwankamol Sastraruji, Stephen G. Pyne, Alison T. Ung, Pitchaya Mungkornawawakul, Wilford Lie, and Araya Jatisatienr

This journal article is available at Research Online: http://ro.uow.edu.au/scipapers/1135
Structural Revision of Stemoburkilline from an E-Alkene to a Z-Alkene

Kwankamol Sastraruji, Stephen G. Pyne, Alison T. Ung, Pitchaya Mungkornasawakul, Wilford Lie, and Araya Jatisaithien

School of Chemistry, University of Wollongong, Wollongong, New South Wales, 2522, Australia, Department of Chemistry, Chiang Mai University, Chiang Mai 50202, Thailand, and Department of Biology, Chiang Mai University, Chiang Mai 50202, Thailand

Received November 27, 2008

Semisynthesis studies starting from (11Z)-1’,2’-didehydrostemofoline (4) that had been isolated earlier, but was not identical. The largest difference was observed for the chemical shift for H-9a, which occurred at δ 3.28 in 8 and was reported to be at δ 3.60 in the natural product. An examination of the original 1H NMR spectra of the original partially purified extracts of Stemona burkillii showed compounds 1, 3, and 8 to be present (42:11:47, respectively; see Supporting Information). In this mixture a signal at δ 3.32 was observed along with other resonances (e.g., δ 5.48 (d, J 10.0 Hz, 1H, H-11) and 4.30 (br s, 1H, H-2)) that were consistent with those of compound 8, however no signal was seen at δ 3.60. Unfortunately we do not have the original sample of stemoburkilline to rerun its 1H NMR spectrum under identical conditions to that of 8. We re-examined the original crude reaction mixture of Stemona burkillii Prain and further NOE studies established stemoburkilline as the Z-isomer (8).

Reports increase steadily each year on the isolation and biological activities of the natural products arising from extracts of plants of Stemona species. Over 100 Stemona alkaloids have been structurally characterized. In 2004 we reported the isolation of two stemofoline alkaloids, 11(S),12(R)-dihydrostemofoline (1) and stemoburkilline (2), along with two known alkaloids from a root extract of Stemona burkillii Prain (Stemonaceae). The structure and relative configuration of 1 were determined via interpretation of its spectroscopic data and from comparison with data from synthetic 11(S),12(S)-dihydrostemofoline (3). The configuration of the exo-cyclic alkene group in 2 was tentively assigned as E on the basis of mechanistic considerations. We had speculated that 2 arose from 1 via a ring-opening reaction involving an elimination process. We report here the synthesis of 1 and 3 from (11Z)-1’,2’-didehydrostemofoline (4) and their base-catalyzed ring-opening reactions to give (Z)-stemoburkilline (8). A re-examination of the crude root extracts of Stemona burkillii Prain and further NOE studies established stemoburkilline as the Z-isomer (8).

MS analysis of the crude reaction mixture. Removal of the TMS ether of 9 under acidic conditions then provided a pure sample of 8 in 61% yield after purification by CC. Under similar conditions compound 1 was converted to 8 in 69% yield (Scheme 2). The 1H NMR spectrum of 8 was similar to that of stemoburkilline, which we had isolated earlier, but was not identical. The largest difference was observed for the chemical shift for H-9a, which occurred at δ 3.28 in 8 and was reported to be at δ 3.60 in the natural product. An examination of the original 1H NMR spectra of the original partially purified extracts of Stemona burkillii showed compounds 1, 3, and 8 to be present (42:11:47, respectively; see Supporting Information). In this mixture a signal at δ 3.32 was observed along with other resonances (e.g., δ 5.48 (d, J 10.0 Hz, 1H, H-11) and 4.30 (br s, 1H, H-2)) that were consistent with those of compound 8, however no signal was seen at δ 3.60. Unfortunately we do not have the original sample of stemoburkilline to rerun its 1H NMR spectrum under identical conditions to that of 8. We re-examined the original crude extracts of S. burkillii that had been kept at −20 °C for 4.5 years. Partial purification of this extract by CC showed compounds 1, 3, and 8 to be present from 1H NMR analysis (36:22:42, respectively; see Supporting Information). In this mixture a signal was observed at δ 3.27, along with those also corresponding to compound 8. With compound 8 in hand we determined it to be...
the Z-isomer on the basis of a NOE cross-peak between the furanone methoxy group and the alkene proton. We suspect that our original NMR sample of stemoburkilline may have had traces of HCl in it, which could have shifted the 'H NMR signals downfield if this were the case. Initial 'H NMR analysis of the mixture (Scheme 1) revealed two major signals for H-9a, although other protons near the protonated nitrogen atom would have also been expected to be observed significantly more downfield if this were the case. Based on these considerations and from the results of our ring-opening experiments, it is quite possible that some of compounds 1, 3, and 8 are artifacts, which have interconverted under nonenzymatic catalyzed reactions either in the plant or during the extraction/purification process. It has proven difficult however to analyze the crude extracts by NMR analysis to determine the ratio of these products due to their relative low abundance.

The stereochemical outcome of the base/TMSCI-initiated ring-opening reaction of 1 and 3 can be rationalized as occurring through an E1cB mechanism, as shown in Scheme 3. Deprotonation of 1 or 3 by DBU at the acidic γ-position of the lactone ring would result in the anionic intermediate A. TMSCI-assisted ring-opening would then give the Z-isomer 9. Ring-opening via the anionic intermediate B, which would lead to (E)-stemoburkilline, would be less likely due to an unfavorable steric interaction between the methoxy and methyl groups in this intermediate (Scheme 3).

Experimental Section

General Experimental Procedures. As described previously, 2,3 'H and 13C NMR assignments were achieved with the aid of gCOSY and, in some cases, NOESY experiments. 13C NMR assignments were based upon DEPT, gHSQC, and gHMBC experiments. All compounds were homogeneous by TLC analysis and judged to be of >95% purity based upon 'H NMR analysis.

Plant Material. The known starting material, (11Z)-1',2'-didehydrostemofoline (4), was isolated from the unidentified Stemona species that we reported earlier. 1 The roots of this Stemona species were collected at Amphur Mae Moh, Lampang, Thailand, in November 2007. The plant material was identified by Mr. James Maxwell as the same species as we had previously studied. 3 A voucher specimen, number 25375, was deposited at the Herbarium of the Department of Biology, Chiang Mai University.

Extraction and Isolation. The dry, ground root of the Stemona species (935 g) was extracted with 95% EtOH (4 x 3000 mL) over 4 days at rt. The ethanolic solution was evaporated to give a dark brown residue (148 g). The extract was partitioned between MeOH/H2O (1:1) and CH2Cl2. The organic extract was dried over MgSO4 and concentrated in vacuo to give a dark brown gum (242.8 mg, 48% w/w).

Stemofoline (5). To a solution of 4 (100.8 mg, 0.262 mmol) in EtOAc (4.0 mL) at rt was added Pd/C (10 mg, 10% w/w), and the flask was flushed with N2 for 10 min before the solution was left to stir under a H2 atmosphere for 1 h. The flask was flushed with N2, and the solution was filtered through Celite and washed with EtOAc. The filtrate was dried over MgSO4 and concentrated in vacuo to give 5 as a yellow-brown gum (98 mg, 0.253 mmol, 96% yield). The NMR data agreed with those reported for the natural product. 4

Isostemofoline (6). To a large NMR tube (5 mm diameter) containing a solution of 5 (48.3 mg, 0.125 mmol) in CHCl3 (2 mL) at rt was added acetonaphthene (50 μL). The mixture was irradiated with a 500 W lamp for 7 h to give a mixture of stemofoline (5) and isostemofoline (6) (ca. 9:1). The mixture was separated by CC using gradient elution from CH2Cl2 to CH2Cl2/MeOH/NH4OH (95:5:1) as eluent to give 6 as a white, amorphous solid (20 mg, 0.052 mmol, 41% yield) and 5 (15.8 mg, 0.041 mmol, 33% yield). The NMR data of 6 agreed with those reported for the natural product. 4
Journal of Natural Products, 2009, Vol. 72, No. 2

11(S),12(S)-Dihydroxystemofoline (3). To a solution of 5 (83.1 mg, 0.214 mmol) in EtOH (3.0 mL) at rt was added NaOH (8.3 mg, 0.10% w/w), and the flask was flushed with N₂ for 10 min before the solution was left to stir under a N₂ atmosphere for 24 h. The flask was flushed with N₂, and the solution was filtered through Celite and washed with MeOH. The filtrate was concentrated in vacuo, and the residue was purified by column chromatography using gradient elution from CH₂Cl₂ to CH₂Cl₂/MeOH/NH₄OH (95:5:1) to give 3 as a colorless gum (20.9 mg, 0.054 mmol, 25% yield) and the ring-opened product 7 (13.5 mg, 0.035 mmol, 16% yield, dr = 72:28) as a brown gum. The NMR data of 3 agreed with that reported."