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# Suffruticosine, a novel octacyclic alkaloid with an unprecedented skeleton from *Securinega suffruticosa* (Pall.) Rehd.

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ABSTRACT

A novel securinega alkaloid, suffruticosine (**1**), was isolated from the leaves and barks of *Securinega suffruticosa*. The structure of **1**, characterized by a fused complex octacyclic system, was elucidated by combined spectroscopic techniques, especially 2D NMR and CD spectral analyses.

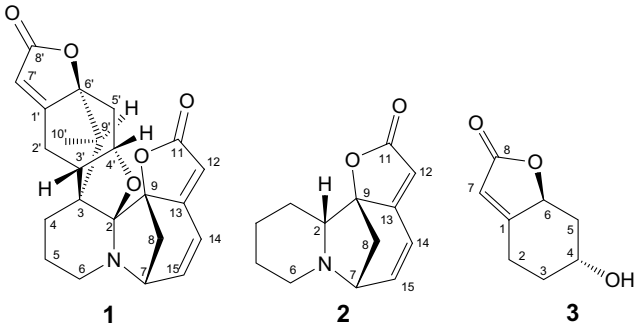


The securinega alkaloids comprise a group of more than 40 tetracyclic compounds produced by some plants of the *Securinega*, *Phyllanthus*, *Margaritaria*, and *Brenia* species, belonging to the Euphorbiaceae family, which have been used for years in traditional folk medicine in China.[1]

*Securinega suffruticosa* (Pall.) Rehd., a kind of semishrub plant widely distributed in temperate zones and subtropical zones, is one of the Chinese folk medicines used to treat rheumatic disease, quadriplegia, impotence, infantile paralysis, children's malnutrition, etc. Previous chemical studies on this plant[2] afforded a number of indolizidine type alkaloids, and some of them showed interesting bioactivities.[3] The complex ring systems and significant biological importance of securinega alkaloids have attracted great interest as challenging targets for total synthesis[4] as well as biosynthetic studies.[5]

In our continuing search for biologically active and structurally unique compounds from Chinese medicinal plants,[6] we have chemically investigated the minor alkaloidal constituents of *S. suffruticosa*, resulting in the isolation of a novel indolizidine alkaloid, suffruticosine (**1**). In this Letter, we describe the isolation and structural elucidation of **1**.

The leaves and barks of *S. suffruticosa* (2.1 kg) collected from Anhui Province, China, were extracted with MeOH. MeOH extracts were partitioned between EtOAc and acidic water (pH 4–5). Water-soluble materials, adjusted to pH 10 with saturated $Na_2CO_3$, were partitioned with $CHCl_3$, and $CHCl_3$-soluble materials were subjected to a silica gel column ($CHCl_3$/MeOH/$Et_2NH$, 99:1:0.1 to 50:50:0.1), in which fractions eluted with $CHCl_3$/MeOH/$Et_2NH$ (95:5:0.1) were purified by an amino silica gel column ($CHCl_3$/MeOH, 99:1 to 90:10) to afford a novel alkaloid, suffruticosine (**1**, 2.1 mg, 0.0001%), together with a known related alkaloid, viroallosecurinine (**2**, 46.2 mg, 0.0023%).[2i]




Suffruticosine (**1**),[7,8] a white amorphous powder, showed the pseudomolecular ion peak at *m*/*z* 394 $(M+H)^+$ in its ESIMS, and its molecular formula, $C_{23}H_{23}NO_5$, was established by HRESIMS [*m*/*z* 416.1483, $(M+Na)^+$, $\Delta$ = +0.9 mmu], indicating 13 degrees of unsaturation. IR absorptions implied the presence of α,β-unsaturated-γ-lactones (1727.9, 1712.5, 1648.9, and 1625.7 $cm^{-1}$), while

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the UV spectrum also indicated the presence of α,β-unsaturated-γ-lactone chromophones ($\lambda_{max}$ 258 nm, log ε 4.06).

The gross structure of **1** was deduced from detailed analysis of $^1$H and $^{13}$C NMR data (Table 1), aided with 2D NMR ($^1$H–$^1$H COSY, TOCSY, HMQC, and HMBC). The NMR data of **1** combined with HMQC experiment indicated that the molecule possessed two ester carbonyl carbons ($\delta_C$ 171.1 and 173.3), two trisubstituted double bonds [($\delta_C$ 111.9, d and 164.3, s; $\delta_H$ 5.91, s), ($\delta_C$ 111.9, d and 164.3, s; $\delta_H$ 5.67, s)], one disubstituted olefin ($\delta_C$ 122.6, d, $\delta_H$ 6.65, d; $\delta_C$ 146.5, d, $\delta_H$ 6.91, dd), three oxygenated sp$^3$ quaternary carbons ($\delta_C$ 85.1, 91.5 and 99.5), one sp$^3$ quaternary carbon ($\delta_C$ 44.5), one sp$^3$ oxymethine ($\delta_C$ 71.5, d, $\delta_H$ 4.43, dd), three sp$^3$ methine ($\delta_C$ 33.7, 38.9 and 56.5), six sp$^3$ methylenes, and one sp$^3$ methyl ($\delta_C$ 9.1, q). Since five of 13 unsaturations were thus accounted for, it was concluded that **1** contained eight rings.

Interpretation of the $^1$H–$^1$H COSY spectrum of **1**, aided by HMQC, TOCSY, and HMBC, revealed the proton connectivities for three partial structures **a**–**c** (Fig. 1).

For the partial structure **a**, the presence of two spin–spin systems ($H_2$-4 to $H_2$-6 and $H_2$-8 to H-14) was evident. Further, the HMBC correlations (Fig. 2) from H-12 ($\delta_H$ 5.91) to C-14, from $H_2$-8 to C-9 ($\delta_C$ 91.5), from $H_2$-4 to C-2 ($\delta_C$ 99.5) and C-3 ($\delta_C$ 44.5), and from H-7 to C-2, C-9, and C-6, bearing in mind four unassigned quaternary carbons (C-2, $\delta_C$ 99.5; C-3, $\delta_C$ 44.5; C-9, $\delta_C$ 91.5; and C-13, $\delta_C$ 164.3), led to unambiguous assignment for the segment. For the partial structure **b**, the oxymethine proton ($\delta_H$ 4.43, H-4′) exhibited clear correlations with the adjacent methylene protons ($\delta_H$ 1.70, H-5′a; $\delta_H$ 2.61, H-5′b) and methine proton ($\delta_H$ 2.56, H-3′), which, in turn, was further correlated with the methylene at $\delta_H$ 2.82 (H-2′a) and $\delta_H$ 2.65 (H-2′b). By analogy to partial structure **a**, the diagnostic HMBC correlations from $H_2$-2′ to C-1′ ($\delta_C$ 169.0) and C-7′ ($\delta_C$ 111.5), and from $H_2$-5′ to C-6′ ($\delta_C$ 85.1) allowed to connect C-1′ to C-2′ and C-5′ to C-6′, according to the segment **b**. Finally, the distinct $^1$H–$^1$H COSY cross-peak between H-9′ ($\delta_H$ 2.30) and $H_3$-10′ ($\delta_H$ 0.52) led to draw the partial structure **c**, being linked to two quaternary carbons, respectively.

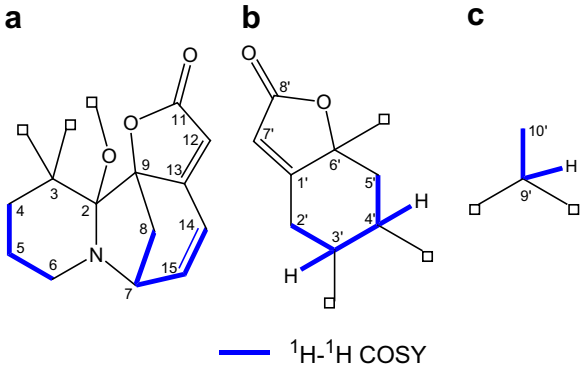


**Figure 1.** Partial structures **a**–**c** of suffruticosine (**1**).

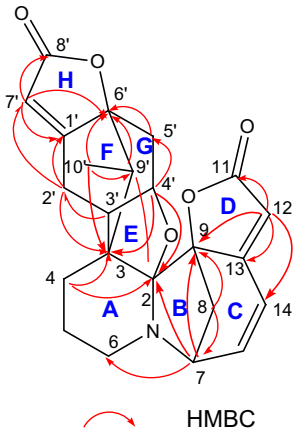


**Figure 2.** Key HMBC correlations of suffruticosine (**1**).

All the subunits were connected together by extensive interpretation of the well-resolved HMBC spectrum. Serious significant $^1$H–$^{13}$C long-range correlations, as shown in Figure 2, linked C-3 to H-3′ and H-9′, C-6′ to H-9′, and C-2 to H-4′. Thus, the planar gross structure of **1** was determined.

Compound **1** contains eight rings (A–H). The relative stereochemistry around the rings E, F, and G was established by detailed analysis of its ROESY spectrum (Fig. 3). The clear NOE correlations between H-4′ and H-3′, H-3′ and H-6a ($\delta_H$ 3.39) indicated that both the rings A and E, rings E and F were all cis-fused, and rings A and F both adopted boat conformation. As a consequence, the bridge involving carbons C-3, C-9′/C-10′, and C-6′ had to be α-oriented. In addition, the ROESY cross-peaks between $H_3$-10′ ($\delta_H$ 0.52) and H-4a ($\delta_H$ 1.55), and between H-9′ ($\delta_H$ 2.30) and H-5′a ($\delta_H$ 2.61) indicated that the configuration of C-9′ was $R^*$.

The absolute configuration of **1** was determined on the basis of the CD spectral analysis (Fig. 4). The CD spectrum of **1** showed a positive Cotton effect at $\lambda_{max}$ 223 nm attributable to the two chromophones of the two α,β-unsaturated-γ-lactones. According to the empirical rule about using the sign of the π–π* Cotton effect to determine the chirality of the γ-carbon of α,β-unsaturated-γ-lactones proposed by Uchida and Kuriyama,[9] the configuration at both C-9 and C-6′ of **1** was tentatively determined *S*.

**Table 1**
$^1$H and $^{13}$C NMR spectral data of compound **1**[a]

| No. | **1** | |
|---|---|---|
| | $\delta_H$ (mult., *J*, Hz)[b] | $\delta_C$[c] |
| 2 | | 99.5, s |
| 3 | | 44.5, s |
| 4a | 1.15 (m) | 20.5, t |
| 4b | 1.55 (m) | |
| 5a | 1.40 (m) | 18.9, t |
| 5b | 1.66 (m) | |
| 6α | 2.91 (dd, 6.0, 3.2) | 39.8, t |
| 6β | 3.39 (dt, 11.6, 3.3) | |
| 7 | 3.79 (t, 2.1) | 56.5, d |
| 8a | 1.73 (m) | 46.3, t |
| 8b | | |
| 9 | | 91.5, s |
| 11 | | 171.0, s |
| 12 | 5.91 (s) | 111.9, d |
| 13 | | 164.3, s |
| 14 | 6.65 (d, 8.8) | 122.6, d |
| 15 | 6.92 (dd, 4.8, 8.8) | 146.5, d |
| 1′ | | 169.0, s |
| 2′α | 2.82 (dd, 12.5, 2.5) | 21.1, t |
| 2′β | 2.65 (dd, 12.5, 2.5) | |
| 3′ | 2.56 (m) | 38.9, d |
| 4′ | 4.43 (ddd, 7.3, 5.1, 2.5) | 71.5, d |
| 5′α | 2.61 (d, 10.1) | 38.7, t |
| 5′β | 1.70 (m) | |
| 6′ | | 85.1, s |
| 7′ | 5.67 (s) | 111.5, d |
| 8′ | | 173.3, s |
| 9′ | 2.30 (q, 7.6) | 33.7, d |
| 10′ | 0.52 (d, 7.4) | 9.1, q |

[a] Bruker DRX; Recorded at 300 MHz and 100 MHz for $^1$H and $^{13}$C, respectively; δ values are reported in ppm referenced to $CDCl_3$ ($\delta_H$ 7.26 and $\delta_C$ 77.0).
[b] The assignments were aided by HMQC, TOCSY, $^1$H–$^1$H COSY, and decoupling experiments.
[c] Deduced by DEPT sequence.

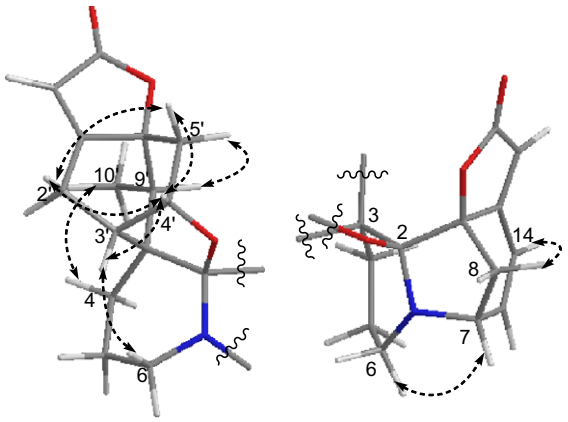


**Figure 3.** Selected ROESY correlations for suffruticosine (**1**).

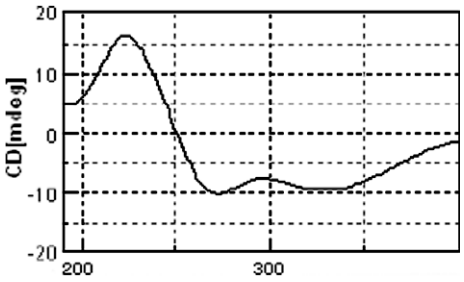


**Figure 4.** CD spectrum of suffruticosine (**1**) $\lambda_{max}$ 223 nm ($\Delta\varepsilon$ + 12.87, MeOH).

To verify the validation of the CD methodology, the CD spectrum of the co-occurring viroallosecurinine (**2**), of which the absolute stereochemistry was known,[10] was also recorded and its expected positive Cotton effect at $\lambda_{max}$ 223 nm is in excellent agreement with the assigned configuration at C-9. Therefore, the absolute configuration of **1** was determined as depicted.

Suffruticosine (**1**) possesses an unprecedented amazing octacyclic ring system consisting of an indoliziding ring, a cyclo-hexene ring, a tetrahydrofuran ring, a seven-membered cyclo-ether ring (G), a cyclo-hexane ring, and two α,β-unsaturated-γ-lactone rings. Although the lower part of the molecule involving the rings A, B, C, and D is closely related to the co-occurring viroallosecurinine (**2**), and the upper half part involving the rings F and H resembling very much (4*R*,6*S*)-2,3-dihydroaquilegiolide (**3**),[11] that was previously isolated from the plant *Sinomenium acutum*, there is no easy way to explain the biogenetic origin of **1**. A hypothetical pathway is tentatively proposed and outlined in Scheme 1 involving the formal oxidative cyclization of **3** and **2** (4′-OH-2; 3′-3) to give the key intermediate structure **4**. Subsequent formal coupling between C-3, C-6′ of **4** and a $C_2$ molecule (e.g., $CH_3CHO$) to form linkages between C-3 and C-9′, and C-9′ and C-6′ should give the octacyclic skeleton of **1**.

The discovery of suffruticosine (**1**) has added to an extremely diverse and complex array of securinega alkaloids. Now, there is a strong interest in performing further studies aimed at experimentally proving the true biogenetic origin and the potential biological role that suffruticosine, and related alkaloids, plays in the life cycle of the plants and finally at confirming their structural peculiarities by synthesis. Compounds **1** and **2** were evaluated for its inhibitory activity against hPTP1B (human protein tyrosine phosphatase 1B), a key target for the treatment of Type-II diabetes and obesity.[12] Unfortunately, the results indicated that both compounds were inactive. Other bioassays, such as antibacterial and anti-inflammatory activities, are currently ongoing.

## Acknowledgments

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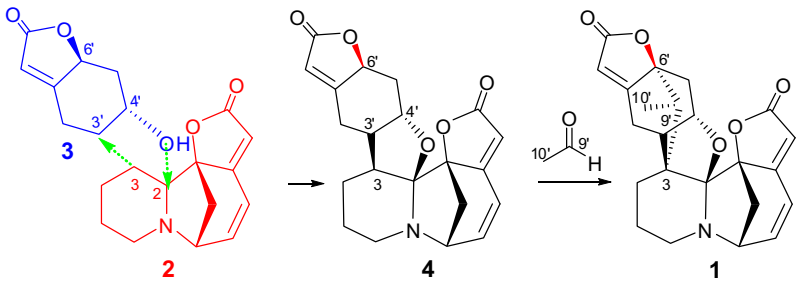
## References and notes

1. An Editorial Committee of the Administration Bureau of Traditional Chinese Medicine. *Chinese Materia Medica* (*Zhonghua Bencao*); Shanghai Science & Technology Press: Shanghai, 1998, pp 856–858.
2. (a) Murav'eva, V. I.; Ban'kovskii, A. I. *Dokl. Akad. Nauk SSSR* **1956**, *110*, 998; (b) Chen, S. F.; Xie, J. X.; Liang, X. T. *Acta Pharm. Sinica* **1963**, *10*, 225–236; (c) Saito, S.; Kotera, K.; Shigematsu, N.; Ide, A.; Sugimoto, N.; Horii, Z.; Hanaoka, M.; Yamawaki, Y.; Tamura, Y. *Tetrahedron* **1963**, *19*, 2085–2099; (d) Horii, Z. H.; Ikeda, M.; Tamura, Y.; Saito, S.; Kotera, K.; Iwamoto, T. *Chem. Pharm. Bull.* **1965**, *13*, 1307–1311; (e) Horri, Z.; Ikeda, M.; Hanaoka, M.; Yamauchi, M.; Tamura, Y.; Saito, S.; Tanaka, T.; Kodera, K.; Sugimoto, N. *Chem. Pharm. Bull.* **1966**, *14*, 917–918; (f) Horri, Z.; Ikeda, M.; Hanaoka, M.; Yamauchi, M.; Tamura, Y.; Saito, S.; Tanaka, T.; Kodera, K.; Sugimoto, N. *Chem. Pharm. Bull.* **1967**, *15*, 1633–1640; (g) Horri, Z.; Yamauchi, M.; Ikeda, M.; Momose, T. *Chem. Pharm. Bull.* **1970**, *18*, 2009–2012; (h) Ohsaki, A.; Ishiyama, H.; Yoneda, K.; Kobayashi, J. *Tetrahedron Lett.* **2003**, *44*, 3097–3099; (i) Wu, H. Y.; Zhou, J. Y. *Chin. J. Chin. Mater. Med.* **2004**, *29*, 535–537; (j) Wang, Y.; Li, Q.; Ye, W.-C.; Ye, C.-F.; Ye, Y.-R.; Zhao, S.-X. *Chin. J. Nat. Med.* **2006**, *4*, 260–263; (k) Wang, Y.; Li, Q.; Ye, W.-C.; Ye, C.-F.; Ye, Y.-R.; Zhao, S.-X. *Chin. Traditional Herbal Drugs* **2007**, *38*, 164–167; (l) Ohsaki, A.; Kobayashi, Y.; Yoneda, K.; Kishida, A.; Ishiyama, H. *J. Nat. Prod.* **2007**, *70*, 2003–2005.
3. (a) Snieckus, V.; Manske, R. E. The *Securinega* Alkaloids. In *The Alkaloids*; Academic Press: New York, 1973; Vol. 14, pp 425–506; (b) Beutler, J. A.; Karbon, E. W.; Brubaker, A. N.; Malik, R.; Curtis, D. R.; Enna, S. J. *Brain Res.* **1985**, *330*, 135–140; (c) Tatematsu, H.; Mori, M.; Yang, T. H.; Chang, J. J.; Lee, T. T. Y.; Lee, K. H. *J. Pharm. Sci.* **1991**, *80*, 325–327; (d) Rognan, D.; Boulanger, T.; Hoffmann, R.; Vercauteren, D. P.; Andre, J. M.; Durant, F.; Wermuth, C. G. *J. Med. Chem.* **1992**, *35*, 1969–1977; (e) Dong, N. Z.; Gu, Z. L. *Acta Pharmacol. Sinica* **1999**, *20*, 267–270.
4. (a) Horii, Z.; Hanoaka, M.; Yamawaki, Y.; Tamura, Y.; Saito, S.; Shigematsu, N.; Kotera, K.; Yoshikawa, H.; Sato, Y.; Nakai, H.; Sugimoto, N. *Tetrahedron* **1967**, *23*, 1165–1174; (b) Heathcock, C. H.; von Geldern, T. W. *Heterocycles* **1987**, *25*, 75–78; (c) Jacobi, P. A.; Blum, C. A.; DeSimone, R. W.; Udodong, U. E. S. *J. Am. Chem. Soc.* **1991**, *113*, 5384–5392; (d) Magnus, P.; Rodríguez-López, J.; Mulholland, K.; Matthews, I. *J. Am. Chem. Soc.* **1992**, *114*, 382–383; (e) Magnus, P.; Rodríguez-López, J.; Mulholland, K.; Matthews, I. *Tetrahedron* **1993**, *49*, 8072–8959; (f) Han, G.; La Porte, M. G.; Folmer, J. J.; Werner, K. M.; Weinreb, S. M. *J. Org. Chem.*




**Scheme 1.** Formal biosynthetic pathway of suffruticosine (**1**).

**2000**, *65*, 6293–6306; (g) Liras, S.; Davoren, J. E.; Bordner, J. *Org. Lett.* **2001**, *3*, 703–706; (h) Honda, T.; Namika, H.; Kudoh, M.; Nagase, H.; Mizutani, H. *Heterocycles* **2003**, *59*, 169–187; (i) Kammler, R.; Polborn, K.; Wanner, K. T. *Tetrahedron* **2003**, *59*, 3359–3368; (j) Alibés, R.; Ballbé, M.; Busqué, F.; de March, P.; Elias, L.; Figueredo, M.; Font, J. *Org. Lett.* **2004**, *6*, 1813–1816; (k) Honda, T.; Namika, H.; Kaneda, K.; Mizutani, H. *Org. Lett.* **2004**, *6*, 87–89; (l) Alibés, R.; Bayón, P.; de March, P.; Figueredo, M.; Font, J.; García-García, E.; González-Gálvez, D. *Org. Lett.* **2005**, *7*, 5107–5109.

5. (a) Sankawa, U.; Yamasaki, K.; Ebizuka, Y. *Tetrahedron Lett.* **1974**, *21*, 1867–1868; (b) Parry, R. J. *J. Chem. Soc., Chem. Commun.* **1975**, 144–145; (c) Golebiewski, W. M.; Horsewood, P.; Spenser, I. D. *J. Chem. Soc., Chem. Commun.* **1976**, 217–218; (d) Sankawa, U.; Ebizuka, Y.; Yamasaki, K. *Phytochemistry* **1977**, *16*, 561–563; (e) Parry, R. J. *Bioorg. Chem.* **1978**, *7*, 277–288; (f) Guan, H.; You, S.; Yang, L.; Wang, X.; Ni, R. *Biotechnol. Lett.* **2005**, *27*, 1189–1193; (g) Yuan, W.; Zhu, P.; Cheng, K.; Meng, C.; Ma, L.; Wu, F.; Zhu, H. X. *Nat. Prod. Res.* **2007**, *21*, 234–242.
6. (a) Sun, Y.-Q.; Guo, Y.-W. *Tetrahedron Lett.* **2004**, *45*, 5533–5535; (b) Zhang, W.; Guo, Y.-W.; Krohn, K. *Chem. Eur. J.* **2006**, *12*, 5122–5127; (c) Li, Z.-Y.; Chen, P.; Xu, H.-G.; Yang, Y.-M.; Peng, S.-Y.; Zhao, Z.-Z.; Guo, Y.-W. *Org. Lett.* **2007**, *9*, 477–480; (d) Wang, J.-D.; Zhang, W.; Li, Z.-Y.; Xiang, W.-S.; Guo, Y.-W. *Phytochemistry* **2007**, *68*, 2426–2431.
7. Suffruticosine (**1**): an amorphous powder; $[\alpha]_D^{20}$ −174 (*c* 0.05, MeOH); UV ($CH_3OH$) $\lambda_{max}$ (log $\varepsilon$) 258 (4.06) nm; IR $\nu_{max}$ (KBr) $cm^{-1}$: 2955, 1727.9, 1712.5, 1648.9, 1625.7, 1461.8 and 1124.3 $cm^{-1}$; ESIMS *m*/*z* 394 $(M+H)^+$; HRESIMS *m*/*z* 416.1483 ($[M+Na]^+$, calcd for $C_{23}H_{23}NO_5Na$, 416.1474); $^1H$ and $^{13}C$ NMR data, see Table 1.
8. Trivial atom numbering by analogy to that of compounds **2** and **3**, respectively.
9. Uchida, I.; Kuriyama, K. *Tetrahedron Lett.* **1974**, *43*, 3761–3764.
10. Nakano, T.; Yang, T. H.; Terao, S. *J. Org. Chem.* **1963**, *28*, 2619–2621.
11. Otsuka, H.; Ito, A.; Fujioka, N.; Kawamata, K. I.; Kasai, R.; Yamasaki, K.; Satoh, T. *Phytochemistry* **1993**, *33*, 389–392.
12. Byon, J. C. H.; Kusari, A. B.; Kuseti, J. *Mol. Cell. Biochem.* **1998**, *182*, 101–108.