# Composite Constituents: Thirty-Nine Triterpenoids Including Two Novel Compounds from *Ixeris chinensis*

Kenji SHIOJIMA,[a] Hideki SUZUKI,[a] Nobuyuki KODERA,[a] Hiroyuki AGETA,*[,a] Hsien-Chang CHANG,[b] and Yuh-Pan CHEN[b]

*Shôwa College of Pharmaceutical Sciences,[a] Machida, Tokyo 194, Japan and Brion Research Institute of Taiwan,[b] 116 Chung-Ching South RD. Sec. 3, Taipei, Taiwan 10743, Republic of China.*
Received September 21, 1995; accepted November 7, 1995

**Thirty-nine triterpenoids including two novel compounds, 17-epilupenyl acetate and ixerenyl acetate, were isolated from the dried aerial parts and roots of *Ixeris chinensis*, Compositae, and their structures were elucidated by spectroscopic analysis.**

**Key words** *Ixeris chinensis*; Compositae; triterpenoid; 17-epilupenyl acetate; ixerenyl acetate

The dried whole plant of *Ixeris chinensis* (THUNB.) NAKAI is a folk medicine in Taiwan with analgesic, antipyretic and anti-inflammatory actions. The isolation of a triterpenoid, bauerenyl acetate, was reported.[1] On reinvestigation of the aerial parts and roots of *I. chinensis*, collected in Taiwan, we have isolated two novel triterpenoids, 17-epilupenyl acetate (**1**)[2] and ixerenyl acetate (**2**),[3] together with thirty-seven known triterpenoids **3**—**39** (Chart 1). This paper deals with the isolation of the above compounds from the acetate, ketone and alcohol fractions, and the structure elucidation of compounds **1** and **2** by means of extensive spectroscopic analysis and chemical correlation.

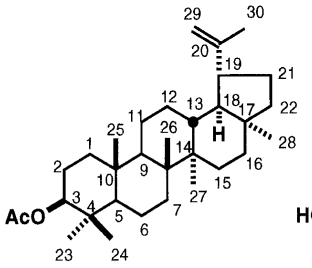
## Results and Discussion

The dried aerial parts and the roots of *Ixeris chinensis* were extracted with hexane, respectively, and extracts were separated by various kinds of chromatography (see Experimental) to give compounds **1**—**39**, which are presented in Table 1 with the physical constants and yields.

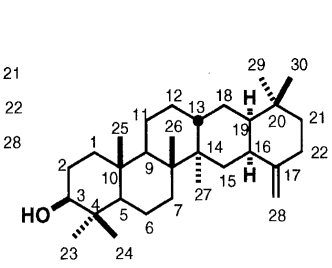
Compound **1** was obtained as colorless plates, and the high-resolution MS (HR-MS) of **1** indicated $M^+$ at *m/z* 468.3965 (Calcd 468.3967), suggesting the molecular formula to be $C_{32}H_{52}O_2$. The IR absorptions of **1** showed the presence of an acetate group. The low-resolution MS (LR-MS) of **1** indicated the base peak at *m/z* 189 (b−AcOH, e) and other major fragment ions at *m/z* (relative intensity): 453 (5, $M^+-CH_3$), 425 (2, $M^+-C_3H_7$), 408 (12, $M^+-AcOH$), 393 (6, $M^+-CH_3-AcOH$), 262 (17, a), 249 (10, b), 218 (11, c), 204 (25, d), 203 (34, d−H), 202 (12, a−AcOH), and 175 (15, c−$C_3H_7$) (Chart 2). This fragmentation pattern was essentially identical with that of **3**.[4] The $^1$H-NMR spectrum of **1** indicated the presence of six tertiary methyl groups, an isopropenyl group, and a 3$\beta$-acetoxyl group in the molecule. The analysis of $^1$H–$^1$H COSY, $^{13}$C–$^1$H COSY, HSQC (heteronuclear single quantum coherence spectroscopy), and HMBC spectra of **1** suggested that **1** was a lupane-type compound, when compared with those of **3**. The partial structure of **1**, shown by heavy lines in Fig. 2, was obtained from the HMBC spectrum. Information on the stereochemistry of **1** was acquired from the NOESY spectrum. That is, NOEs were observed between H-24 and H-25, H-25 and H-26, H-26 and H-13$\beta$, H-13$\beta$ and H-19$\beta$; H-5$\alpha$ and H-9$\alpha$, H-9$\alpha$ and H-27, H-27 and H-28; and H-27, H-28 and H-18$\alpha$ (Table 2 and Fig. 1). The structure of rings A, B, C and D of **1** was the same as that of **3**, while the D and E ring juncture was determined by the *cis* configuration of 18$\alpha$-H and 28$\alpha$-methyl. Two chair–chair–chair–boat–envelope conformations of **1** with different side chain form (**1′**, shown in Fig. 1, steric energy 82.394 kcal/mol, and **1″**, whose side chain at C-19 is in the opposite orientation, 82.810 kcal/mol) were simulated by Chem3D Plus/MM2.[5] The preferred conformation **1′** was well supported by NOEs as shown in the figure, especially H-30 and H-28. The presence of another preferred conformation **1″** in solution was proved by the two singlet signals of H-29 protons and NOEs between H-29b and H-18$\alpha$; H-30 and H-19$\beta$. This case is very similar to that of hop-22(29)-ene.[6] Although the yield of **1** was only 1/75 of that of **3**, the former could be a very interesting alternative product of lupeol biosynthesis.

The alcohol fraction was acetylated with $Ac_2O$–pyridine, and the mixture was isolated by various kinds of chromatography to give ixerenyl acetate (**2a**) (see Experimental). The HR-MS of **2a** indicated $M^+$ at *m/z* 468.3959 (Calcd 468.3967), suggesting the molecular formula to be $C_{32}H_{52}O_2$. The IR absorption of **2a** showed the presence of an acetate group. The LR-MS of **2a** indicated the base peak at *m/z* 189 (b, k) and major fragment ions at *m/z* (relative intensity): 453 (9, $M^+-CH_3$), 425 (5, f), 408 (32, $M^+-AcOH$), 393 (25, $M^+-CH_3-AcOH$), 365 (4, f−AcOH), 262 (25, a), 249 (12, b), 218 (31, g, h), 204 (61, i, j), 202 (24, a−AcOH), and 135 (35, l) (Chart 2). This fragmentation pattern indicated that the structure of rings A, B and C of **2a** was essentially identical with that of **3**.[4] The $^1$H-NMR spectrum of **2a** showed the presence of seven tertiary methyl groups, an exocyclic methylene group, and a 3$\beta$-acetoxyl group in the molecule. The $^1$H- and $^{13}$C-signals of rings A, B and C (except C-12 and C-13 in **2a**) were very similar to those of **3**, suggesting the same structure (Tables 2 and 3). The analysis of the $^1$H–$^1$H COSY, HSQC, and HMBC spectra suggested that **2a** had a methylene (C-18) between C-13 and C-19, a methine (C-16), and an exocyclic methylene (C-28) attached to C-17. The partial structure of **2a**, shown by heavy lines in Fig. 2, was obtained from the HMBC spectrum. In addition, all methylene and methine carbons

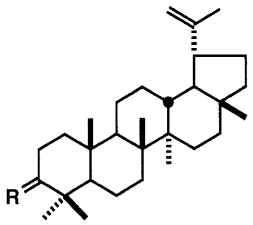
* To whom correspondence should be addressed.



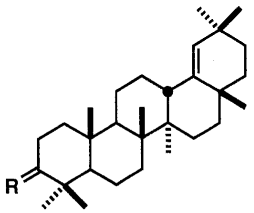

1



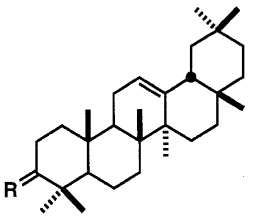

2



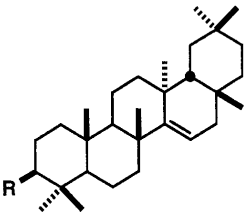

**3** R=αH, βOAc
**15** R=O
**22** R=αH, βOH



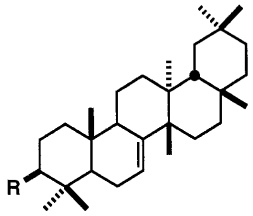

**4** R=αH, βOAc
**16** R=O
**23** R=αH, βOH



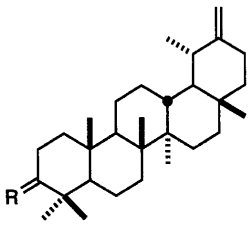

**5** R=αH, βOAc
**17** R=O
**24** R=αH, βOH



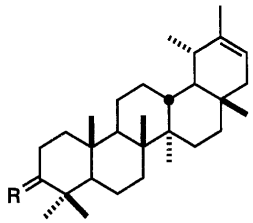

**6** R=OAc
**26** R=OH



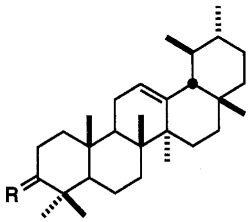

**7** R=OAc
**27** R=OH



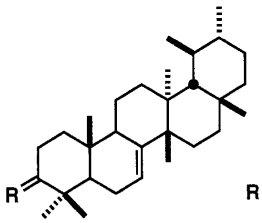

**8** R=αH, βOAc
**18** R=O
**28** R=αH, βOH



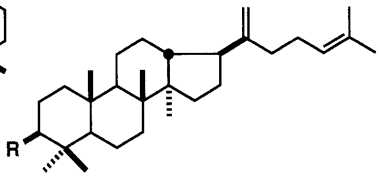

**9** R=αH, βOAc
**19** R=O
**29** R=αH, βOH



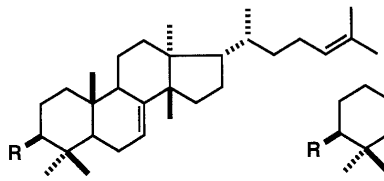

**10** R=αH, βOAc
**20** R=O
**30** R=αH, βOH



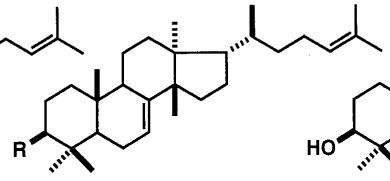

**11** R=αH, βOAc
**21** R=O
**31** R=αH, βOH



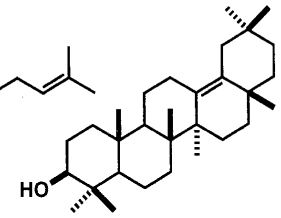

**12** R=OAc
**33** R=OH



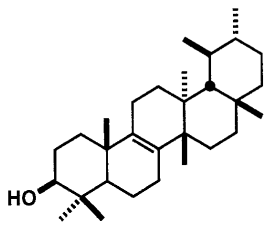

**13** R=OAc
**34** R=OH



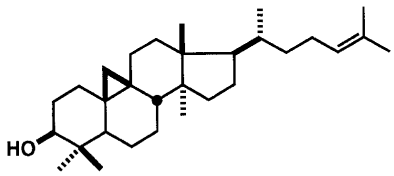

**14** R=OAc
**35** R=OH



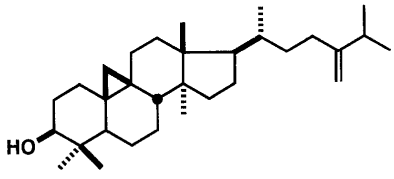

**25**



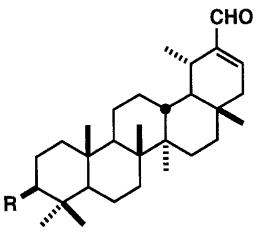

**32**




**36**




**37**




**38** R=OAc
**39** R=OH

Chart 1

were correlated from the corresponding proton signals, confirmed by the $^{1}H$–$^{1}H$ COSY spectrum with the signals of proton(s) attached to the neighboring carbon(s).[6] Information on the stereochemistry of **2a** was obtained by NOE spectrometry. That is, NOEs were observed between H-24 and 25, H-25 and 26, H-26 and 13$\beta$, H-13$\beta$

Table 1. Triterpenoids Isolated from *Ixeris chinensis*

| | mp (°C) | $[\alpha]_D$ (°) | Yield[a] (%) Aerial parts | Yield[a] (%) Roots | Ref. |
|---|---|---|---|---|---|
| 17-Epilupenyl acetate (**1**) | 219—221 | +24.7 | 0.0009 | 0.0006 | 2 |
| Ixerenol (**2**) | | | 0.0006 | | 3 |
| Lupenyl acetate (**3**) | 221—222 | | 0.0667 | 0.0449 | 7 |
| Germanicyl acetate (**4**) | 280—282 | | 0.0302 | 0.0196 | 7 |
| β-Amyrin acetate (**5**) | 241—243 | | 0.0404 | 0.0455 | 7 |
| Taraxeryl acetate (**6**) | 281—283 | | | Trace | 10 |
| Multiflorenyl acetate (**7**) | 214—215 | | 0.0069 | 0.0030 | 7 |
| Taraxasteryl acetate (**8**) | 240—242 | | 0.0944 | 0.0764 | 7 |
| ψ-Taraxasteryl acetate (**9**) | 238—240 | | 0.0640 | 0.0659 | 7 |
| α-Amyrin acetate (**10**) | 223—224 | | 0.0311 | 0.0494 | 7 |
| Bauerenyl acetate (**11**) | 294—296 | | 0.0141 | 0.0278 | 7 |
| Dammaradienyl acetate (**12**) | 146—147 | +61.2 | 0.0050 | | 8 |
| Butyrospermyl acetate (**13**) | 140—142 | | 0.0015 | 0.0016 | 7 |
| Tirucalla-7,21-dien-3β-yl acetate (**14**) | 115—117 | | 0.0059 | 0.0026 | 7 |
| Lupenone (**15**) | | | Trace | 0.0014 | 7 |
| Germanicone (**16**) | | | Trace | 0.0007 | 7 |
| β-Amyrenone (**17**) | | | Trace | 0.0020 | 7 |
| Taraxasterone (**18**) | | | Trace | 0.0030 | 7 |
| ψ-Taraxasterone (**19**) | | | Trace | 0.0033 | 7 |
| α-Amyrenone (**20**) | | | Trace | 0.0018 | 7 |
| Bauerenone (**21**) | | | Trace | 0.0003 | 7 |
| Lupeol (**22**) | | | 0.0185 | 0.0032 | 7 |
| Germanicol (**23**) | | | 0.0004 | 0.0012 | 7 |
| β-Amyrin (**24**) | | | 0.0009 | 0.0051 | 7 |
| Olean-13(18)-en-3β-ol (**25**) | | | | 0.009 | 13 |
| Taraxerol (**26**) | | | Trace | 0.0003 | 10 |
| Multiflorenol (**27**) | | | | Trace | 7 |
| Taraxasterol (**28**) | | | 0.0290 | 0.0087 | 7 |
| ψ-Taraxasterol (**29**) | | | 0.0218 | 0.0126 | 7 |
| α-Amyrin (**30**) | | | 0.0011 | 0.0041 | 7 |
| Bauerenol (**31**) | | | Trace | 0.0004 | 7 |
| Isobauerenol (**32**) | | | | 0.007 | 7 |
| Dammaradienol (**33**) | | | | Trace | 8 |
| Butyrospermol (**34**) | | | 0.0023 | Trace | 7 |
| Tirucalla-7,21-dien-3β-ol (**35**) | | | 0.0006 | Trace | 7 |
| Cycloartenol (**36**) | | | 0.0003 | | 11 |
| 24-Methylene-cycloartenol (**37**) | | | 0.0001 | 0.0008 | 12 |
| 3β-Acetoxytaraxaster-20-en-30-al (**38**) | 230—232 | +84.8 | 0.0034 | 0.0023 | 9 |
| 3β-Hydroxytaraxaster-20-en-30-al (**39**) | | | Trace | 0.0003 | 9 |

a) Yield from the dried materials after removal of water by azeotropic distillation.

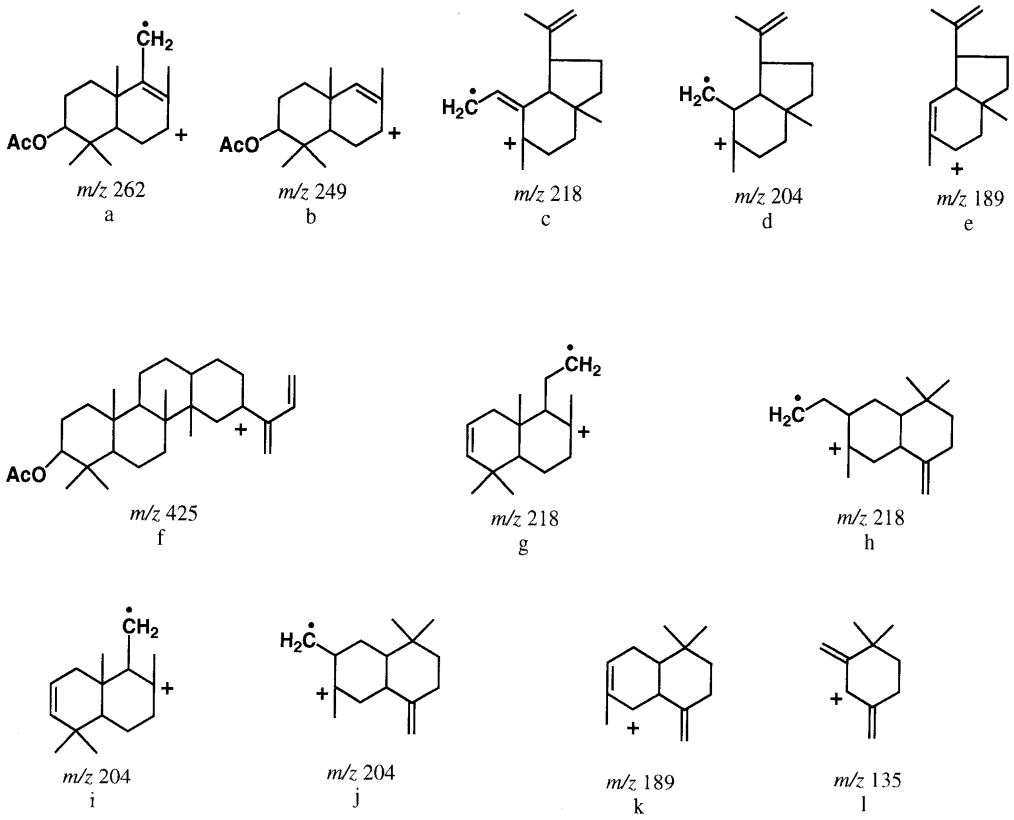



Chart 2

and 30, H-15β and 30, H-15β and 22β, H-21β and 30, H-22β and 30; H-9α and 27, H-27 and 16α, H-16α and 19α, H-16α and 28a, H-19α and 29, and H-22α and 28b (Fig. 1). The D and E ring juncture was determined by the *cis* configuration of 16α-H and 19α-H. Therefore, **2a** was established to have a new type of skeleton, as shown in Chart 1. Biogenetically, we suggest that cyclization of squalene oxide gives the germanicane cation, whose C-17

Table 2. $^1$H-NMR Spectral Data (500 MHz, $CDCl_3$, $\delta$)

| | H-1 | H-2 | H-3 | H-5 | H-6 | H-7 | H-9 | H-11 |
|---|---|---|---|---|---|---|---|---|
| **1** | 1.01; 1.70 | 1.64; 1.64 | 4.477 (dd, 5.8, 10.8) | 0.80 | 1.52; 1.36 | 1.33; 1.33 | 1.33 | 1.48; 1.18 |
| **2a** | 1.02; 1.71 | 1.63; 1.63 | 4.481 (dd, 5.8, 10.9) | 0.81 | 1.50; 1.37 | 1.37; 1.37 | 1.39 | 1.47; 1.28 |
| **3** | 1.00; 1.67 | 1.62; 1.62 | 4.471 (dd, 5.8, 10.7) | 0.80 | 1.48; 1.40 | 1.38; 1.38 | 1.32 | 1.40; 1.31 |

| | H-12 | H-13 | H-15 | H-16 | H-18 | H-19 | H-21 | H-22 |
|---|---|---|---|---|---|---|---|---|
| **1** | 0.88; 1.58 | 1.19 | 1.07; 1.50 | 1.45; 1.51 | 1.35 | 2.272 (ddd, 3.1, 8.9, 8.9) | 1.53; 1.84 | 1.74; 1.14 |
| **2a** | 1.00; 1.33 | 1.81 | 0.85; 1.79 | 2.50 | 1.31; 1.48 | 1.46 | 1.26; 1.45 | 2.02; 2.31 |
| **3** | 1.08; 1.67 | 1.66 | 1.00; 1.68 | 1.36; 1.45 | 1.36 | 2.376 (ddd, 5.8, 11.2, 11.2) | 1.32; 1.92 | 1.37; 1.19 |

| | H-23 | H-24 | H-25 | H-26 | H-27 | H-28 | H-29 | H-30 |
|---|---|---|---|---|---|---|---|---|
| **1** | 0.848 | 0.837 | 0.865 | 0.953 | 0.865 | 0.901 | a 4.638 (br s) b 4.741 (br s) | 1.734 |
| **2a** | 0.844 | 0.838 | 0.876 | 0.981 | 0.954 | 4.576 4.579 | 0.892 | 1.087 |
| **3** | 0.845 | 0.835 | 0.854 | 1.029 | 0.938 | 0.786 | a 4.568 (m) b 4.686 (br d, 2.5) | 1.683 |

Coupling constants are shown in parentheses and acetyl methyl protons were observed at $\delta$ 2.043 in **1**, $\delta$ 2.044 in **2a**, $\delta$ 2.041 in **3**. Methylene signals are shown for $\alpha$; $\beta$.

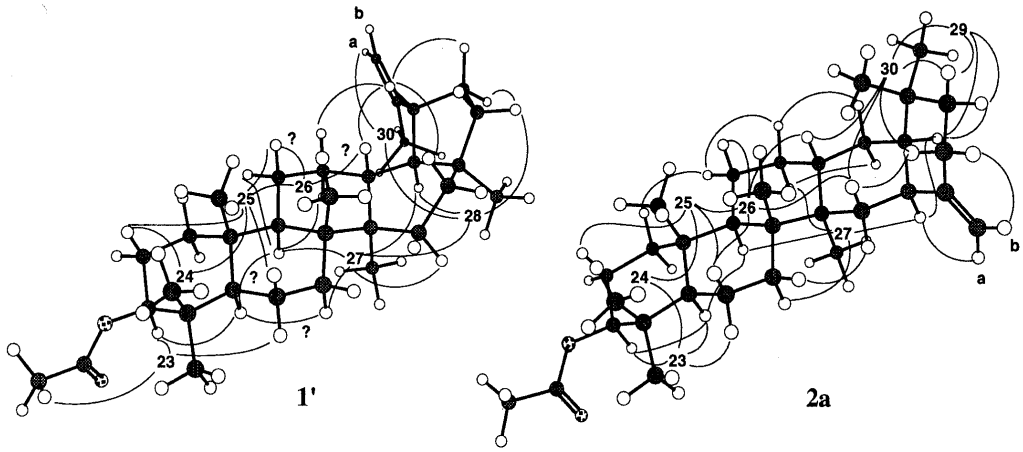



Fig. 1. Chem3D Plus Drawing and NOEs of **1'** and **2a**

and C-18 bond open, followed by recyclization to afford **2** (Chart 3).

The triterpenoids obtained from the aerial parts and roots of *Ixeris chinensis* are mainly pentacyclic triterpenoids belonging to the lupane, oleanane, migrated oleanane, ursane, and migrated ursane groups, with some tetracyclic compounds.

**Experimental**

Melting points were measured on a Yanagimoto micro apparatus without correction. Specific rotations were observed in $CHCl_3$ solution ($c$=0.1) at 22—24 °C. The $^1$H- and $^{13}$C-NMR spectra were taken at 500 and 270/125 MHz, respectively, by the Fourier-transform (FT) method in $CDCl_3$ solution with tetramethylsilane as an internal standard. MS was recorded (direct inlet) at 30 eV and the relative intensities of peaks were reported with reference to the most intense peak higher than $m/z$ 100. GC was run on a Hitachi 163 apparatus using a glass column containing Chromosorb G HP coated with SE-30 (1.4%) at 260 °C in a flow of $N_2$. Cholestane was used as an internal reference, and its retention time was set at 3.0 min. GC-MS was run on a JGC 20K-JMS D300 system using the same absorbant as described above in a flow of He. HPLC was performed on a C-18 reverse-phase column (8 i.d. × 250 mm, refractive index detector) with $CH_3CN$–$CHCl_3$ (9 : 1, 9.5 : 0.5) as the eluent. Silica gel 60, 230—400 mesh (Merck), and 20% $AgNO_3$–impregnated silica gel were used for column chromatography (CC).

**Plant Material** The aerial parts and roots of *Ixeris chinensis* were collected in January, 1993, at Tao-yuan, Taiwan. Voucher specimens have been deposited in the Brion Research Institute of Taiwan.

**Extraction of Dried Aerial Parts of *I. chinensis* and Separation** The dried aerial parts (1.7 kg) were extracted three times with hexane. The extract was evaporated and the residue (60.3 g) was chromatographed on silica gel with hexane (frs. A, B), hexane–benzene (8 : 2) (frs. C—E), hexane–benzene (1 : 1) (fr. F), benzene (frs. G, H), benzene–ether (9 : 1) (frs. I, J) and ether (frs. K, L) to give twelve fractions.

**17-Epilupenyl Acetate (1), Lupenyl Acetate (3), Germanicyl Acetate (4), $\beta$-Amyrin Acetate (5), Multiflorenyl Acetate (7), Taraxasteryl Acetate (8), $\psi$-Taraxasteryl Acetate (9), $\alpha$-Amyrin Acetate (10), Bauerenyl Acetate (11), Dammaradienyl Acetate (12), Butyrospermyl Acetate (13), Tirucalla-7,21-dien-3$\beta$-yl Acetate (14) and 3$\beta$-Acetoxytaraxaster-20-en-**

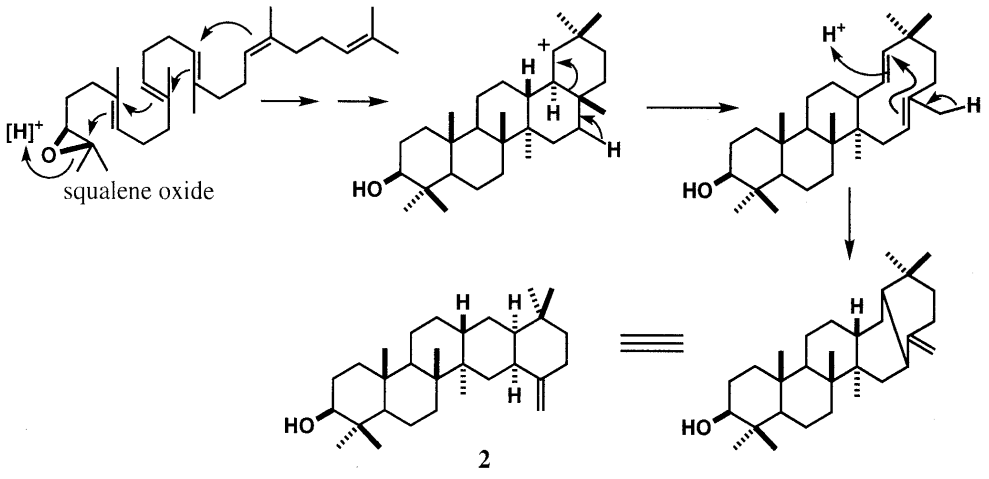
Table 3. $^{13}$C-NMR Spectral Data (125 MHz, $CDCl_3$, $\delta$)

| | Carbon numbers | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
| **1** | 38.57 | 23.72 | 80.98 | 37.83 | 55.65 | 18.13 | 33.79 | 40.87 | 51.33 | 37.16 |
| **2a** | 38.62 | 23.71 | 80.97 | 37.84 | 55.66 | 18.06 | 33.74 | 40.77 | 51.24 | 37.23 |
| **3** | 38.38 | 23.71 | 80.97 | 37.79 | 55.37 | 18.20 | 34.20 | 40.85 | 50.34 | 37.08 |

| | 11 | 12 | 13 | 14 | 15 | 16 | 17 | 18 | 19 | 20 |
|---|---|---|---|---|---|---|---|---|---|---|
| **1** | 21.57 | 27.07 | 43.36 | 40.66 | 27.33 | 33.08 | 40.47 | 49.39 | 54.16 | 150.94 |
| **2a** | 21.37 | 30.25 | 33.17 | 40.90 | 36.45 | 40.94 | 155.44 | 29.58 | 43.30 | 33.77 |
| **3** | 20.94 | 25.10 | 38.04 | 42.82 | 27.42 | 35.57 | 43.00 | 48.28 | 48.00 | 150.97 |

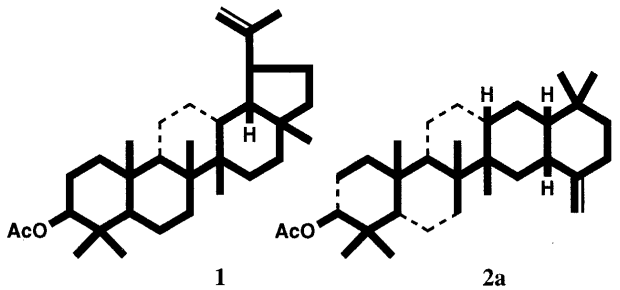
| | 21 | 22 | 23 | 24 | 25 | 26 | 27 | 28 | 29 | 30 |
|---|---|---|---|---|---|---|---|---|---|---|
| **1** | 29.62 | 37.46 | 27.92 | 16.50 | 16.61 | 15.60 | 14.84 | 29.99 | 107.63 | 22.60 |
| **2a** | 44.18 | 28.66 | 27.92 | 16.49 | 16.56 | 15.56 | 14.30 | 106.47 | 32.59 | 25.83 |
| **3** | 29.83 | 39.99 | 27.95 | 16.50 | 16.17 | 15.97 | 14.51 | 18.00 | 109.35 | 19.28 |

Acetyl signals were observed at $\delta$ 21.33, 171.04 in **1**, $\delta$ 21.34, 171.04 in **2a**, $\delta$ 21.32, 171.03 in **3**.




**2**

Chart 3




**1** **2a**

Fig. 2. Partial Structures of **1** and **2a**, Based on the HMBC Spectrum

**30-al (38)** Fraction E was chromatographed on 20% $AgNO_3$–impregnated silica gel with hexane–benzene (8 : 2) followed by HPLC with $CH_3CN$–$CHCl_3$ (9 : 1, 9.5 : 0.5) to give the following crystalline solids (recrystallized from acetone to obtain pure specimens). **1**, 14 mg, IR $\nu_{max}^{KBr}$ cm$^{-1}$: 1732, 1248. **3**, 1.02 g. **4**, 464 mg, $^1$H-NMR ($\delta$): 0.847 (H-23), 0.847 (H-24), 0.904 (H-25), 1.077 (H-26), 0.732 (H-27), 1.017 (H-28), 0.936 (H-29), 0.936 (H-30), 4.483 (dd, $J$=7.1, 9.1 Hz, H-3), 4.862 (H-19), 2.046 (–OCOC$\underline{H}_3$). **5**, 621 mg, $^1$H-NMR ($\delta$): 0.874 (H-23), 0.874 (H-24), 0.966 (H-25), 0.966 (H-26), 1.129 (H-27), 0.829 (H-28), 0.874 (H-29), 0.874 (H-30), 4.505 (dd, $J$=7.3, 8.7 Hz, H-3), 5.181(dd, $J$=3.7, 3.7 Hz, H-12), 2.058 (–OCOC$\underline{H}_3$). **7**, 106 mg, $^1$H-NMR ($\delta$): 0.857 (H-23), 0.936 (H-24), 0.765 (H-25), 1.077 (H-26), 1.071 (H-27), 1.058 (H-28), 0.970 (H-29), 0.970 (H-30), 4.503 (dd, $J$=5.9, 7.7 Hz, H-3), 5.466 (ddd, $J$=3.0, 3.0, 3.4 Hz, H-7), 2.055 (–OCOC$\underline{H}_3$). **8**, 1.45 g, $^1$H-NMR ($\delta$): 0.852 (H-23), 0.852 (H-24), 0.875 (H-25), 1.018 (H-26), 0.926 (H-27), 0.852 (H-28), 1.018 (d, $J$=6.7 Hz, H-29), 4.608 (m, H-30), 4.484 (dd, $J$=6.9, 8.1 Hz, H-3), 2.047 (–OCOC$\underline{H}_3$). **9**, 982 mg, $^1$H-NMR ($\delta$): 0.852 (H-23), 0.852 (H-24), 0.876 (H-25), 1.043 (H-26), 0.945 (H-27), 0.734 (H-28), 0.989 (d, $J$=6.4 Hz, H-29), 1.633 (H-30), 4.488 (dd, $J$=6.1, 9.3 Hz, H-3), 5.261 (br d, $J$=6.7 Hz, H-21), 2.049 (–OCOC$\underline{H}_3$). **10**, 478 mg, $^1$H-NMR ($\delta$): 0.866 (H-23), 0.866 (H-24), 0.977 (H-25), 1.009 (H-26), 1.065 (H-27), 0.798 (H-28), 0.796 (d, $J$=5.8 Hz, H-29), 0.913 (br s, H-30), 4.505 (dd, $J$=7.3, 8.7 Hz, H-3), 5.123 (dd, $J$=3.5, 3.5 Hz, H-12), 2.051 (–OCOC$\underline{H}_3$). **11**, 217 mg, $^1$H-NMR ($\delta$): 0.848 (H-23), 0.933 (H-24), 0.770 (H-25), 0.994 (H-26), 0.943 (H-27), 1.038 (H-28), 1.032 (d, $J$=6.7 Hz, H-29), 0.904 (d, $J$=6.1 Hz, H-30), 4.514 (dd, $J$=6.7, 8.5 Hz, H-3), 5.410 (ddd, $J$=3.2, 3.2, 3.7 Hz, H-7), 2.055 (–OCOC$\underline{H}_3$). **12**, 76 mg, $^1$H–NMR ($\delta$): 0.852 (H-23), 0.852 (H-24), 0.852 (H-25), 0.972 (H-26), 0.865 (H-27), 4.708, 4.737 (H-28), 1.617 (H-29), 1.688 (H-30), 5.133 (m, H-21), 4.483 (dd, $J$=6.6, 9.1 Hz, H-3), 2.047 (–OCOC$\underline{H}_3$). **13**, 22 mg, $^1$H-NMR ($\delta$): 0.851 (H-23), 0.933 (H-24), 0.764 (H-25), 0.973 (H-26), 0.802 (H-27), 0.847 (d, $J$=6.1 Hz, H-28), 1.606 (H-29), 1.688 (H-30), 4.514 (dd, $J$=6.7, 8.5 Hz, H-3), 5.247 (ddd, $J$=3.1, 3.1, 3.7 Hz, H-7), 5.098 (m, H-21), 2.057 (–OCOC$\underline{H}_3$). **14**, 91 mg, $^1$H-NMR ($\delta$): 0.851 (H-23), 0.933 (H-24), 0.766 (H-25), 0.967 (H-26), 0.805 (H-27), 0.882 (d, $J$=6.1 Hz, H-28), 1.605 (H-29), 1.686 (H-30), 4.515 (dd, $J$=6.1, 9.1 Hz, H-3), 5.248 (ddd, $J$=3.1, 3.1, 3.7 Hz, H-7), 5.099 (m, H-21), 2.057 (–OCOC$\underline{H}_3$). **38**, 51 mg, MS $m/z$ (rel. int): 482.3776 ($M^+$, 5), 422 (67), 407 (28), 249 (8), 232 (3), 230 (6), 218 (11), 217 (14), 216 (10), 203 (31), 189 (100), $^1$H-NMR ($\delta$): 0.854 (H-23), 0.845 (H-24), 0.879 (H-25), 1.034

(H-26), 0.966 (H-27), 0.670 (H-28), 1.021 (d, $J$=6.4 Hz, H-29), 9.368 (H-30, –CHO), 4.488 (dd, $J$=6.3, 10.3 Hz, H-3), 6.711 (dd, $J$=2.7, 6.7 Hz, H-21), 2.048 (–OCOCH$_3$), $^{13}$C-NMR ($\delta$): 38.44 (C-1), 23.69 (C-2), 80.94 (C-3), 37.80 (C-4), 55.40 (C-5), 18.17 (C-6), 34.14 (C-7), 41.05 (C-8), 50.30 (C-9), 37.03 (C-10), 21.45 (C-11), 27.25 (C-12), 39.01 (C-13), 42.27 (C-14), 26.88 (C-15), 36.48 (C-16), 34.80 (C-17), 48.21 (C-18), 29.39 (C-19), 148.45 (C-20), 149.20 (C-21), 43.02 (C-22), 27.95 (C-23), 16.51 (C-24), 16.34 (C-25), 15.99 (C-26), 14.69 (C-27), 17.52 (C-28), 23.17 (C-29), 194.05 (C-30, –CHO), 21.32 (–OCOCH$_3$), 171.05 (–OCOCH$_3$). Compounds **3**—**14**, **38** were identified by comparison of their melting point, MS, $^1$H-NMR and $^{13}$C-NMR data with those of authentic samples or published values.[7-9]

**Lupenone (15), Germanicone (16), β-Amyrenone (17), Taraxasterone (18), ψ-Taraxasterone (19), α-Amyrenone (20) and Bauerenone (21)** Fraction F was chromatographed on silica gel. The crystalline solids from the hexane–benzene (1 : 1) eluates were identified by comparison of their GC R$t_R$ and GC-MS data with those of authentic samples.[4,7] **15**, GC and GC-MS: R$t_R$ 2.91, $m/z$: 424, 409, 381, 205, 203, 189. **16**, GC and GC-MS: R$t_R$ 2.69, $m/z$: 424, 409, 205, 204, 203, 189, 177. **17**, GC and GC-MS: R$t_R$ 2.64, $m/z$: 424, 409, 218, 205, 203, 189. **18**, GC and GC-MS: R$t_R$ 3.52, $m/z$: 424, 409, 218, 205, 204, 203, 189. **19**, GC and GC-MS: R$t_R$ 3.43, $m/z$: 424, 409, 218, 205, 204, 203, 189. **20**, GC and GC-MS: R$t_R$ 2.87, $m/z$: 424, 409, 218, 205, 204, 203, 189. **21**, GC and GC-MS: R$t_R$ 3.50, $m/z$: 424, 409, 257, 245, 205, 189, 177.

**Ixerenol (2), Lupeol (22), Germanicol (23), β-Amyrin (24), Taraxerol (26), Taraxasterol (28), ψ-Taraxasterol (29), α-Amyrin (30), Bauerenol (31), Dammaradienol (33), Butyrospermol (34), Tirucalla-7,21-dien-3β-ol (35), Cycloartenol (36), 24-Methylenecycloartanol (37), and 3β-Hydroxytaraxaster-20-en-30-al (39)** Fraction G was acetylated with acetic anhydride–pyridine. This acetate mixture was purified and identified by the same method as used in the case of fraction E (acetate fraction). Ixerenyl acetate (**2a**), 9 mg, mp 158—160 °C, $[\alpha]_D$ +39.5°, IR $\nu_{max}^{KBr}$ cm$^{-1}$: 1732, 1248. Lupenyl acetate (**22a**), 284 mg, mp 216—218 °C. Germanicyl acetate (**23a**), 5.4 mg. β-Amyrin acetate (**24a**), 14 mg, mp 240—242 °C. Taraxeryl acetate (**26a**), 0.3 mg, $^1$H-NMR ($\delta$): 0.860 (H-23), 0.875 (H-24), 0.948 (H-25), 1.091 (H-26), 0.948 (H-27), 0.820 (H-28), 0.907 (H-29), 0.907 (H-30), 4.462 (dd, $J$=6.8, 9.0 Hz, H-3), 5.532 (dd, $J$=3.2, 8.1 Hz, H-15), 2.045 (–OCOCH$_3$). Taraxasteryl acetate (**28a**), 445 mg, mp 239—242 °C. ψ-Taraxasteryl acetate (**29a**), 335 mg, mp 238—239 °C. α-Amyrin acetate (**30a**), 17 mg, mp 222—224 °C. Bauerenyl acetate (**31a**), 0.6 mg. Dammaradienyl acetate (**33a**), 27 mg, mp 138—140 °C. Butyrospermyl acetate (**34a**), 36 mg, mp 138—140 °C. Tirucalla-7,21-dien-3β-yl acetate (**35a**), 9 mg, mp 114—116 °C. Cycloartenyl acetate (**36a**), 5 mg, mp 121—122 °C, MS $m/z$: 468 ($M^+$), 453, 408, 393, 297, 295, 286, 69, $^1$H-NMR ($\delta$): 0.960 (H-18), 0.341, 0.573 (d, $J$=4.3 Hz, H-19), 0.881 (d, $J$=6.4 Hz, H-21), 1.685 (H-26), 1.607 (H-27), 0.846 (H-30), 0.889 (H-31), 0.889 (H-32), 4.564 (dd, $J$=5.2, 10.7 Hz, H-3), 5.101 (m, H-24), 2.055 (–OCOCH$_3$). 24-Methylenecycloartanyl acetate (**37a**), 2 mg, mp 112—113 °C, $^1$H-NMR ($\delta$): 0.963 (H-18), 0.343, 0.576 (d, $J$=4.3 Hz, H-19), 0.894 (d, $J$=6.1 Hz, H-21), 1.025 (d, $J$=6.7 Hz, H-26), 1.030 (d, $J$=6.7 Hz, H-27), 4.664, 4.717 (H-28), 0.889 (H-30), 0.901 (H-31), 0.847 (H-32), 4.566 (dd, $J$=4.7, 11.1 Hz, H-3), 2.056 (–OCOCH$_3$). 3β-Hydroxytaraxaster-20-en-30-al (**39a**), trace. Compounds **22a**—**37a**, **39a** were identified by comparison of their melting point and $^1$H-NMR and MS data with those of authentic samples or the published values.[7-12]

**Extraction of Dried Roots of *I. chinensis* and Separation** The dried roots (454 g) were extracted three times with hexane. The extract was evaporated and the residue (5.9 g) was chromatographed on silica gel with hexane (fr. A′), hexane–benzene (8 : 2) (fr. B′), hexane–benzene (7 : 3) (fr. C′), hexane–benzene (6 : 4) (fr. D′), hexane–benzene (1 : 1) (frs. E′—G′), benzene (frs. H′, I′), benzene–ether (9 : 1) (frs. J′, K′), and ether (frs. L′, M′) to give thirteen fractions.

**17-Epilupenyl Acetate (1), Lupenyl Acetate (3), Germanicyl Acetate (4), β-Amyrin Acetate (5), Taraxeryl Acetate (6), Multiflorenyl Acetate (7), Taraxasteryl Acetate (8), ψ-Taraxasteryl Acetate (9), α-Amyrin Acetate (10), Bauerenyl Acetate (11), Butyrospermyl Acetate (13), Tirucalla-7,21-dien-3β-yl Acetate (14) and 3β-Acetoxytaraxaster-20-en-30-al (38)** Fraction D′ was chromatographed on 20% $AgNO_3$–impregnated silica gel with hexane–benzene (8 : 2) followed by HPLC with $CH_3CN$–$CHCl_3$ (9 : 1) to give the following crystalline solids (recrystallized from acetone to obtain pure specimens): **1**: 2 mg; **3**: 204 mg; **4**: 89 mg; **5**: 207 mg; **6**: trace; **7**: 13 mg; **8**: 347 mg; **9**: 299 mg; **10**: 224 mg; **11**: 129 mg; **13**: 7 mg; **14**: 12 mg; **38**: 11 mg. Compounds **3**—**14**, **38** were identified by comparison of their melting point and $^1$H-NMR data with those of authentic samples or the published values.[7,9,10]

**Lupenone (15), Germanicone (16), β-Amyrenone (17), Taraxasterone (18), ψ-Taraxasterone (19), α-Amyrenone (20), Bauerenone (21)** Fraction F′ was chromatographed repeatedly on 20% $AgNO_3$–impregnated silica gel with hexane–benzene (1 : 1) to give seven fractions. These fractions were identified by comparison of their $^1$H-NMR data with those of authentic samples.[7] **15**, 6 mg, $^1$H-NMR ($\delta$): 1.070 (H-23), 1.024 (H-24), 0.934 (H-25), 1.074 (H-26), 0.955 (H-27), 0.799 (H-28), 4.573, 4.692 (H-29), 1.684 (H-30). **16**, $^1$H-NMR ($\delta$): 1.081 (H-23), 1.034 (H-24), 0.965 (H-25), 1.106 (H-26), 0.746 (H-27), 1.028 (H-28), 0.944 (H-29), 0.944 (H-30), 4.863 (H-19). **17**, $^1$H-NMR ($\delta$): 1.097 (H-23), 1.057 (H-24), 1.076 (H-25), 1.023 (H-26), 1.145 (H-27), 0.842 (H-28), 0.874 (H-29), 0.874 (H-30), 5.208 (dd, $J$=3.5, 3.5 Hz, H-12). **18**, $^1$H-NMR ($\delta$): 1.079 (H-23), 1.034 (H-24), 0.952 (H-25), 1.057 (H-26), 0.944 (H-27), 0.866 (H-28), 1.022 (d, $J$=6.4 Hz, H-29), 4.618 (m, H-30). **19**, $^1$H-NMR ($\delta$): 1.079 (H-23), 1.034 (H-24), 0.952 (H-25), 1.079 (H-26), 0.965 (H-27), 0.746 (H-28), 0.990 (d, $J$=6.4 Hz, H-29), 1.638 (H-30), 5.266 (br d, $J$=6.7 Hz, H-21). **20**, $^1$H-NMR ($\delta$): 1.097 (H-23), 1.057 (H-24), 1.080 (H-25), 1.057 (H-26), 1.081 (H-27), 0.811 (H-28), 0.798 (d, $J$=6.7 Hz, H-29), 0.914 (br s, H-30), 5.153 (dd, $J$=3.7, 3.7 Hz, H-12). **21**, $^1$H-NMR ($\delta$): 1.110 (H-23), 1.044 (H-24), 1.044 (H-25), 1.002 (H-26), 0.943 (H-27), 1.044 (H-28), 1.042 (d, H-29), 0.904 (d, H-30), 5.468 (ddd, H-7).

**Lupeol (22), Germanicol (23), β-Amyrin (24), Olean-13(18)-en-3β-ol (25), Taraxerol (26), Multiflorenol (27), Taraxasterol (28), ψ-Taraxasterol (29), α-Amyrin (30), Bauerenol (31), Isobauerenol (32), Dammaradienol (33), Butyrospermol (34), Tirucalla-7,21-dien-3β-ol (35), 24-Methylenecycloartanol (37), and 3β-Hydroxytaraxaster-20-en-30-al (38)** Fraction G′ was acetylated with acetic anhydride–pyridine. This acetate mixture was purified and identified by the same method as used for fraction E (acetate fraction). Lupenyl acetate (**22a**), 15 mg, mp 217—218 °C. Germanicyl acetate (**23a**), 5 mg. β-Amyrin acetate (**24a**), 23 mg, mp 238—240 °C. Olean-13(18)-en-3β-yl acetate (**25a**), 4 mg, $^1$H-NMR ($\delta$): 0.852 (H-23), 0.838 (H-24), 0.881 (H-25), 0.864 (H-26), 1.152 (H-27), 1.005 (H-28), 0.697 (H-29), 0.933 (H-30), 4.505 (dd, $J$=7.0, 9.3 Hz, H-3), 2.050 (–OCOCH$_3$). Taraxeryl acetate (**26a**), 1 mg. Multiflorenyl acetate (**27a**), trace. Taraxasteryl acetate (**28a**), 40 mg, mp 238—240 °C. ψ-Taraxasteryl acetate (**29a**), 57 mg, mp 237—238 °C. α-Amyrin acetate (**30a**), 19 mg, mp 221—223 °C. Bauerenyl acetate (**31a**), 2 mg. Isobauerenyl acetate (**32a**), 3 mg. Dammaradienyl acetate (**33a**), trace. Butyrospermyl acetate (**34a**), trace. Tirucalla-7,21-dien-3β-yl acetate (**35a**), trace. 24-Methylenecycloartanyl acetate (**37a**), 3 mg. 3β-Acetoxytaraxaster-20-en-30-al (**39a**), 2 mg. Compounds **22a**—**35a**, **37a**, **39a** were identified by comparison of their melting point and $^1$H-NMR data with those of authentic samples or the published values.[7-13]

**Acknowledgement** The authors are indebted to Mr. Yôichi Takase of this College for MS measurements.

**References and Notes**

1) Huang Y. L., Chen C. C., Chen Y. P., Hsu H. Y., *J. Taiwan Pharm.*, **36**, 211 (1984).
2) Shiojima K., Suzuki H., Kodera N., Kubota K., Tsushima S., Ageta H., Chang H. C., Chen Y. P., *Chem. Pharm. Bull.*, **42**, 2193 (1994).
3) Shiojima K., Suzuki H., Kodera N., Ageta H., Chang H. C., Chen Y. P., *Chem. Pharm. Bull.*, **43**, 180 (1995).
4) Shiojima K., Arai Y., Masuda K., Takase Y., Ageta T., Ageta H., *Chem. Pharm. Bull.*, **40**, 683 (1992).
5) Chem3D Plus 3.0 and N. L. Allinger, MM2 Params, Cambridge Scientific Computing Inc.
6) Ageta H., Shiojima K., Suzuki H., Nakamura S., *Chem. Pharm. Bull.*, **41**, 1939 (1993).
7) Shiojima K., Masuda K., Suzuki H., Lin T., Ooishi Y., Ageta H., *Chem. Pharm. Bull.*, **43**, 1634 (1995).
8) Talapatra S. K., Bhar D. S., Talapatra B., *Aust. J. Chem.*, **27**, 1137 (1974).
9) Herz W., Kulanthaivel P., *Phytochemistry*, **22**, 513 (1983).
10) Matsunaga S., Tanaka R., Akagi M., *Phytochemistry*, **27**, 535 (1988).
11) Arai Y., Masuda K., Ageta H., *Shoyakugaku Zasshi*, **38**, 53 (1984).
12) Ageta H., Arai Y., *Phytochemistry*, **23**, 2875 (1984).
13) Tanaka R., Matsunaga S., *Phytochemistry*, **27**, 3579 (1988).