# Fern Constituents: *Adiantum cuneatum*. III. Four New Triterpenoids, 4,23-Bisnor-3,4-secofilic-5(24)-en-3-al, 4,23-Bisnor-3,3-dimethoxy-3,4-secofilic-5(24)-ene, 7β,25-Epoxyfern-9(11)-en-8α-ol and 7α,8α-Epoxyfernan-25-ol

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**Four new triterpenoids, 4,23-bisnor-3,4-secofilic-5(24)-en-3-al (1), 4,23-bisnor-3,3-dimethoxy-3,4-secofilic-5(24)-ene (2), 7β,25-epoxyfern-9(11)-en-8α-ol (3) and 7α,8α-epoxyfernan-25-ol (4) were isolated from the fresh leaves of *Adiantum cuneatum*, and their structures were elucidated by means of spectroscopic analysis.**

**Key words** fern; *Adiantum cuneatum*; triterpenoid; secofilicane group; fernane group

In the preceding paper of this series,[1] we reported the isolation and structure elucidation of nine new compounds, as well as thirteen known triterpenoids, from the fresh leaves of *Adiantum cuneatum* LANGSD. and FISCH. (*A. raddianum* PRESL, Adiantaceae). Further fractionation of the same extract resulted in the isolation of four more new triterpenoids *viz.* 4,23-bisnor-3,4-secofilic-5(24)-en-3-al (**1**),[2] 4,23-bisnor-3,3-dimethoxy-3,4-secofilic-5(24)-ene (**2**),[2] 7β,25-epoxyfern-9(11)-en-8α-ol (**3**) and 7α,8α-epoxyfernan-25-ol (**4**) along with three known triterpenoids. This paper deals with structure elucidation of **1**—**4**.

## Results and Discussion

The more polar fractions than those reported earlier[1a] were purified by various chromatographic techniques (see Experimental) to afford four new triterpenoids **1**—**4** together with three known compounds **5**—**7**, which are summarized in Table 1 along with their physical constants and yields.

Compound **1** and **2** were obtained as colorless needles and plates, respectively. The IR spectrum of **1** indicated the presence of a carbonyl group, whereas that of **2** showed neither carbonyl nor hydroxyl absorptions. The high-resolution mass spectra (HR-MS) of **1** and **2** showed their molecular formulae to be $C_{28}H_{46}O$ ($M^+$ *m/z* 398.3558, Calcd 398.3548) and $C_{30}H_{50}O_2$ ($M^+$ *m/z* 444.3979, Calcd 442.3811), respectively. Their low-resolution MS (LR-MS) showed the same fragment ions at *m/z* 355 (a), 287 (b), 274 (base peak, c), 259 (c-15), 205 (d) and 191(e) (Chart 2)[3] with different intensities, suggesting that both compounds have almost the same partial structure or skeleton. Their $^1$H-NMR (Table 2) spectra displayed signals due to four tertiary and two secondary methyl groups and an exocyclic methylene group; the signals of H-26—H-30 were very similar. The signals of an aldehyde proton in **1** and two methoxyl groups in **2** were observed, and the $^{13}$C chemical shifts (Table 3) of C-1, C-2 and C-3 in the two compounds were very different from each other.

A detailed analysis of the heteronuclear multiple bond correlation (HMBC) spectra of **1** and **2** clearly revealed the same carbon skeleton, shown by heavy lines in Fig. 1, thereby indicating that both compounds possess 4,23-bisnor-3,4-secofilic-5(24)-ene structure with a difference at C-3. The relative configurations of the stereogenic centers of **1** and **2** were established by nuclear Overhauser effect spectroscopy (NOESY), in which NOE interactions were observed between methyl, methylene and methine groups situated on the α side: (H-10—H-8—$H_3$-27—$H_3$-28) and on the β side ($H_2$-24—$H_3$-25—$H_3$-26), respectively. Thus, the structures of compounds **1** and **2** were established to be 4,23-bisnor-3,4-secofilic-5(24)-en-3-al and 4,23-bisnor-3,3-dimethoxy-3,4-secofilic-5(24)-ene, respectively.

Compound **3** was obtained as colorless needles and its IR spectrum suggested the presence of a hydroxyl group in the molecule. Its molecular formula was found to be $C_{30}H_{48}O_2$ by HR-MS ($M^+$ *m/z* 440.3701, Calcd 440.3654). The $^1$H-NMR spectrum of **3** indicated the presence of five tertiary methyl and two secondary methyl groups, and a carbinyl methylene (Table 2). Since $^1$H- and $^{13}$C-NMR data (Tables 1 and 2) were not conclusive for determination of the structure of the compound, the HMBC spectrum of **3** was recorded. Detailed analysis of the HMBC (partial structure shown in Fig. 1) revealed that the compound is a fern-9(11)-ene derivative with an ether linkage between C-25 and C-7. The $^{13}$C chemical shift of δ 84.08 suggested that a hydroxyl group was

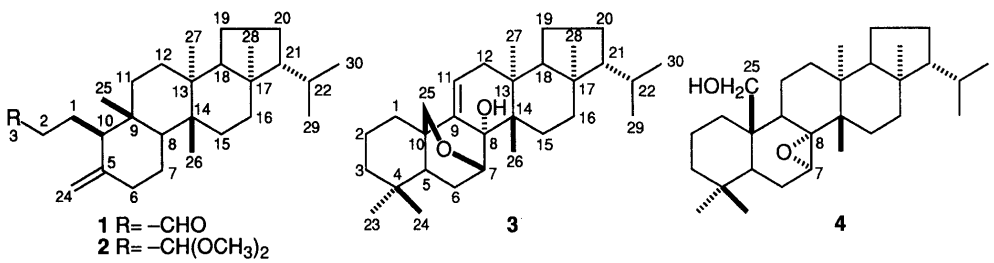


Chart 1

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attached to C-8. This is also supported by the comparatively deshielded chemical shift of $H_3$-27 in comparison with that of fern-9(11)-ene.[4] The relative stereochemistry at most of the chiral centers of **3** was established by the NOESY spectrum as depicted in Fig. 2. Thus, the structure of **3** was determined as 7$\beta$,25-epoxyfern-9(11)-en-8$\alpha$-ol.

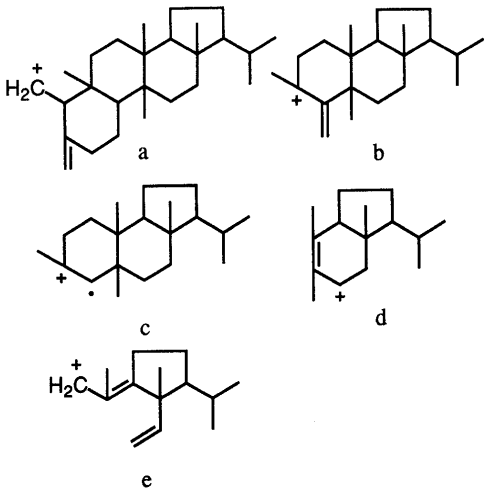


Chart 2

Compound **4** was obtained as colorless needles and its IR spectrum suggested the presence of a hydroxyl group in the molecule. Its molecular formula was deduced to be $C_{30}H_{50}O_2$ by HR-MS ($M^+$ $m/z$ 442.3803, Calcd 442.3811). The $^1$H-NMR spectrum of **4** indicated the presence of five tertiary and two secondary methyl groups, a hydroxy methylene group, and a carbinyl methine group (Table 2). Although the $^1$H- and $^{13}$C-NMR data (Tables 2 and 3) were not conclusive for determination of the structure of **4**, the HMBC spectrum clearly revealed the presence of the partial structure showed by heavy lines in Fig. 1. The presence of a 7$\alpha$,8$\alpha$-epoxide in **4** was also indicated by the down-field shift of $H_3$-27 ($\delta$ 1.087, $\alpha$-Me) in comparison with that of fern-7-en-25-ol.[1b] The relative stereochemistry at most of the chiral centers of **4** was established by the NOESY spectrum, which showed NOE interactions between methyl, methylene and methine protons on the $\beta$-side ($H_3$-24—$H_2$-25—$H_3$-26—H-18—H-21, H-7—H-26), and on the $\alpha$-side ($H_3$-27—$H_3$-28). Thus, the structure of **4** was determined as 7$\alpha$,8$\alpha$-epoxyfernan-25-ol.

This is the first time that secotriterpenoids such as **1** and **2** have been found in fern plants. A possible biogenetic pathway for **1** and **2** is shown in Chart 3. The double bond at C-3–C-4 in filic-3-ene (**8**)[1a] is cleaved oxidatively; then

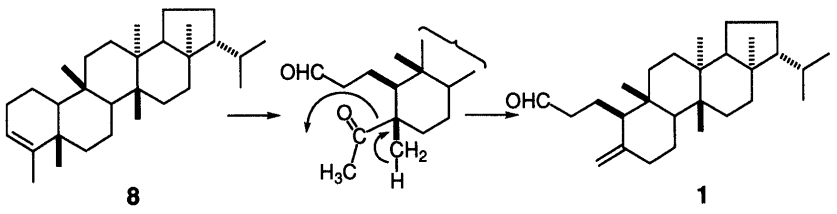


Chart 3

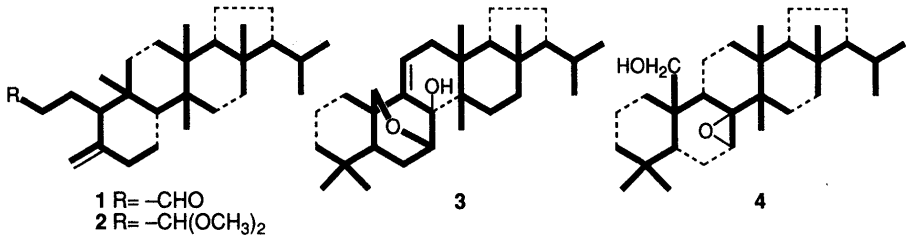


Fig. 1. Partial Structures of **1**, **2**, **3** and **4**, Based on the HMBC Spectra

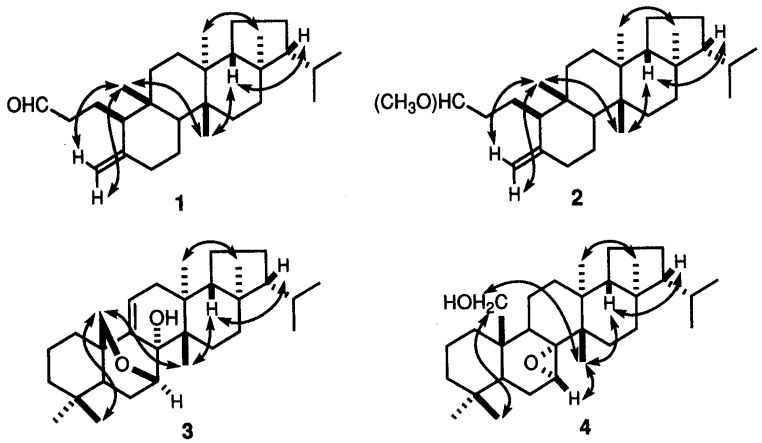


Fig. 2. NOEs Obtained from NOESY

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Table 1. Triterpenoids Isolated from *A. cuneatum*

| | mp (°C) | $[\alpha]_D^{23}$ (°) | Yield[a] (%) | Ref. |
|---|---|---|---|---|
| 4,23-Bisnor-3,4-secofilic-5(24)-en-3-al (**1**) | 138—139 | +0.9 | 0.0003 | 2 |
| 4,23-Bisnor-3,3-dimethoxy-3,4-secofilic-5(24)-ene (**2**) | 195—196 | −5.4 | 0.0015 | 2 |
| 7β,25-Epoxyfern-9(11)-en-8α-ol (**3**) | 229—230 | −59.0 | 0.0002 | |
| 7α,8α-Epoxyfernan-25-ol (**4**) | 250—251 | −49.6 | 0.0024 | |
| Hydroxyhopane (**5**) | 253—255 | +44.0 | 0.0001 | 5 |
| Ketohakonanol (**6**) | 295—297 | +8.0 | 0.0002 | 5 |
| Isoadiantol B (**7**) | 213.5—215.0 | +16.0 | 0.0829 | 5 |

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

Table 2. $^1$H-NMR Spectral Data[a] (500 MHz, $CDCl_3$, δ)

| | **1** | **2** | **3** | **4** |
|---|---|---|---|---|
| H-23 | — | — | 0.903 | 0.883 |
| H-24 | 4.422 (ddd, 1.5, 1.5, 1.2) 4.813 (ddd, 1.5, 1.5, 1.2) | 4.505 (ddd, 1.5, 1.5, 1.5) 4.795 (ddd, 1.5, 1.5, 1.5) | 0.935 | 0.893 |
| H-25 | 0.712 | 0.690 | 2.975 (d, 7.6) 4.185 (d, 7.6) | 3.484 (d, 7.9) 4.052 (d, 7.9) |
| H-26 | 0.881 | 0.875 | 0.979 | 1.109 |
| H-27 | 0.974 | 0.973 | 1.121 | 1.087 |
| H-28 | 0.788 | 0.787 | 0.790 | 0.751 |
| H-29 | 0.825 (d, 6.4) | 0.824 (d, 6.4) | 0.904 (d, 6.4) | 0.887 (d, 6.4) |
| H-30 | 0.881 (d, 6.4) | 0.882 (d, 6.4) | 0.834 (d, 6.4) | 0.822 (d, 6.4) |
| H-7β | | | | 4.068 (dd, 5.5, 5.5) |
| H-11 | | | 5.424 (dd, 4.6, 2.4) | |

Multiplicity and coupling constants (*J*, Hz) are shown in parentheses. *a*) Assignments have been done on the basis of DEPT, $^1$H–$^1$H COSY, $^1$H–$^{13}$C COSY, HMBC and NOESY spectra.

a methyl ketone group is eliminated from the intermediate seco-compound and the exomethylene moiety is formed. *A. cuneatum* also contains compounds having a fernane skeleton with an unusual ether linkage between C-25 and C-7, *viz.* 7β,25-epoxyfern-8-ene[1a] and 7β,25-epoxyfern-9(11)-en-8α-ol(**3**).

## Experimental

General procedure and the plant material: see the preceding papers.[1]

**4,23-Bisnor-3,4-secofilic-5(24)-en-3-al (1), 4,23-Bisnor-3,3-dimethoxy-3,4-secofilic-5(24)-ene (2), 7β,25-Epoxyfern-9(11)-en-8α-ol (3) and 7α,8α-Epoxyfernan-25-ol (4), Hydroxyhopane (5), Ketohakonanol (6), and Isoadiantol B (7)** Fractions E and F (see the preceding paper[1a]) were chromatographed on silica gel with benzene followed by HPLC with $CH_3CN$–$CHCl_3$ (19 : 1) to give the following crystalline solids (recrystallized from acetone or $CHCl_3$–MeOH to give pure compounds).

**1**, 3 mg, IR $\nu_{max}^{KBr}$ cm$^{-1}$: 1700, MS *m/z* (rel. int.): 398 (17, $M^+$), 383 (20, $M^+-15$), 355 (24, a), 287 (35, b), 274 (100, c), 259 (61, c-15), 205 (43, d) and 191 (67, e). **2**, 15 mg, MS *m/z* (rel. int.): 444 (2, $M^+$), 429 (3, $M^+-15$), 354 (48, a-H), 287 (50, b), 274 (100, c), 259 (66, c-15), 205 (51, d) and 191 (48, e). **3**, 3 mg, IR $\nu_{max}^{KBr}$ cm$^{-1}$: 3547, 1080, MS *m/z* (rel. int.): 440 (72), 425 (25), 412 (29), 397 (100), 356 (7), 287 (2), 205 (51) and 191 (10). **4**, 23 mg, IR $\nu_{max}^{KBr}$ cm$^{-1}$: 3440, 1069, MS *m/z* (rel. int.): 442 (100), 427 (52). **5**, 1 mg. **6**, 2 mg. **7**, 482 mg. Compounds **5**, **6** and **7** were identified by direct comparison (IR, $^1$H-NMR) with authentic samples.[5]

Table 3. $^{13}$C-NMR Spectral Data[a] (125 MHz, $CDCl_3$, δ)

| | **1** | **2** | **3** | **4** |
|---|---|---|---|---|
| C-1 | 15.79 | 31.55 | 30.73 | 30.78 |
| C-2 | 43.22 | 18.45 | 20.19 | 20.42 |
| C-3 | 203.05 | 105.10 | 42.17 | 42.05 |
| C-4 | — | — | 33.31 | 33.25 |
| C-5 | 147.95 | 148.44 | 44.60 | 47.23 |
| C-6 | 38.04 | 38.14 | 32.69 | 32.39 |
| C-7 | 23.72 | 23.74 | 71.47 | 69.81 |
| C-8 | 48.78 | 48.80 | 84.08 | 87.55 |
| C-9 | 39.19 | 39.06 | 143.37 | 47.85 |
| C-10 | 57.20 | 57.74 | 45.78 | 47.78 |
| C-11 | 33.64 | 33.70 | 117.14 | 15.93 |
| C-12 | 29.05 | 29.11 | 37.34 | 31.67 |
| C-13 | 38.82 | 38.81 | 38.50 | 37.35 |
| C-14 | 40.36 | 40.36 | 44.28 | 44.34 |
| C-15 | 29.36 | 29.37 | 26.31 | 28.05 |
| C-16 | 35.64 | 35.69 | 35.53 | 35.58 |
| C-17 | 42.71 | 42.72 | 43.02 | 42.72 |
| C-18 | 51.72 | 51.75 | 52.85 | 54.74 |
| C-19 | 19.96 | 19.96 | 20.54 | 20.44 |
| C-20 | 28.38 | 28.39 | 28.16 | 28.21 |
| C-21 | 60.07 | 60.08 | 59.53 | 59.51 |
| C-22 | 30.77 | 30.78 | 30.80 | 30.75 |
| C-23 | — | — | 32.41 | 32.61 |
| C-24 | 106.40 | 106.32 | 21.87 | 21.61 |
| C-25 | 18.09 | 18.19 | 74.48 | 71.83 |
| C-26 | 16.06 | 16.05 | 18.69 | 19.91 |
| C-27 | 15.41 | 15.34 | 21.64 | 23.07 |
| C-28 | 16.23 | 16.22 | 14.58 | 14.70 |
| C-29 | 21.95 | 21.95 | 22.97 | 22.96 |
| C-30 | 22.90 | 22.91 | 22.14 | 22.09 |

*a*) Assignments have been done on the basis of DEPT, $^1$H–$^1$H COSY, $^1$H–$^{13}$C COSY and HMBC spectra.

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