NMR Spectra of Triterpenoids. II. Hopenes and Migrated Hopenes

Hiroyuki AGETA,* Kenji SHIOJIMA, Yōko ARAI, Hideki SUZUKI, and Tamiko KIYOTANI

Shōwa College of Pharmaceutical Sciences, Higashi-Tamagawagakuen, Machida, Tokyo 194, Japan.
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The ^{1}H- and ^{13}C-NMR signals of eleven triterpenoid hydrocarbons belonging to the hopene and migrated hopene groups were completely assigned by the application of modern NMR techniques, and the conformations are discussed on the basis of Chem3D Plus and MM2 calculations, and nuclear Overhauser effect spectroscopy spectra.

Keywords ^{1}H-NMR; ^{13}C-NMR; triterpenoid; hopene; migrated hopene; conformation

We have investigated many kinds of hopane and migrated hopane triterpenoids, mainly isolated from ferns, since 1959. ^{1}H-NMR and ^{13}C-NMR spectra of the compounds have been very useful for their structural determination. In earlier days, assignments of NMR signals were made mostly by analogy with related compounds and were ambiguous in some cases. Recent developments in NMR measurement of organic compounds mean that precise assignments of ^{1}H- and ^{13}C-signals can now be made without comparison with those of related compounds in most cases. In a previous paper[1] we reported the side chain conformation and the precise assignments of NMR signals of eight triterpenoids belonging to the hopane and isohopane groups, including hop-22(29)-ene (**1**). This paper deals with the assignments of the ^{1}H- and ^{13}C-NMR signals of eleven triterpenoid hydrocarbons belonging to the hopane and migrated hopane groups: hop-21-ene (**2**),[2] hop-17(21)-ene (**3**),[2] hop-16-ene (**4**),[3] neohop-13(18)-ene (**5**),[4] neohop-12-ene (**6**),[4] pteron-14-ene (**7**),[5] fern-9(11)-ene (**8**),[6] fern-8-ene (**9**),[7] fern-7-ene (**10**),[7] adian-5-ene (**11**),[6,8] and filic-3-ene (**12**),[6] with consideration of minimum-steric-energy conformations simulated by using the Chem3D Plus and MM2 programs.[9] The assignments of the H-29, H-30, and C-29, C-30 signals of these compounds were firmly established for the first time.

Experimental

General Procedure ^{1}H-/^{13}C-NMR spectra of the compounds in $CDCl_3$ solution were run at 500/125 MHz. The ^{13}C-signals were classified by means of the distortionless enhancement by polarization transfer (DEPT) method and the signals were correlated with ^{1}H-signals by the ^{13}C–^{1}H correlated spectroscopy (C–H-COSY) method. Methyl proton signals and related carbon signals were correlated by the heteronuclear multiple bond correlation (HMBC) method. The assignments of methyl signals determined by the $CDCl_3$–C_6H_6 solvent shift method[2,10] were confirmed by the HMBC spectra. Signals of methylene and methine protons were picked up from the C–H-COSY spectra, and linkages to the same or the neighboring carbons were made by the ^{1}H–^{1}H correlated spectroscopy (H–H-COSY) method. On the other hand, the most stable conformation of each compound with minimum steric energy was simulated by using the Chem3D Plus and MM2 programs[9] and finally the conformation was confirmed by obtaining nuclear Overhauser effect spectroscopy (NOESY) spectra. Signals of methylene and methine protons were also confirmed by the NOESY spectra including the stereochemistry of the methylene protons.

Numbering of the Side Chain Numbering of the isopropyl side chains is shown in Chart 1, following the system of neriifoliol and dryocrassol.[11] In the case of compounds **3**—**12**, C-29 is *pro R* and C-30 is *pro S*.

Conditions for NMR Measurement The 1D and 2D NMR spectra were measured on a JEOL A500 spectrometer equipped with a VAX station 3200 computer, using a $CDCl_3$ solution of *ca.* 10 mg in 0.8 ml with tetramethylsilane (TMS) as the internal standard, at room temperature (24 °C). The chemical shifts are reported on the δ scale. For 1D ^{1}H-NMR at 500.00 MHz, 16 K data points and a frequency width of 10000.0 Hz were used, giving a digital resolution of 0.6 Hz per point. For 1D ^{13}C-NMR at 125.65 MHz, 16 K data points and a frequency

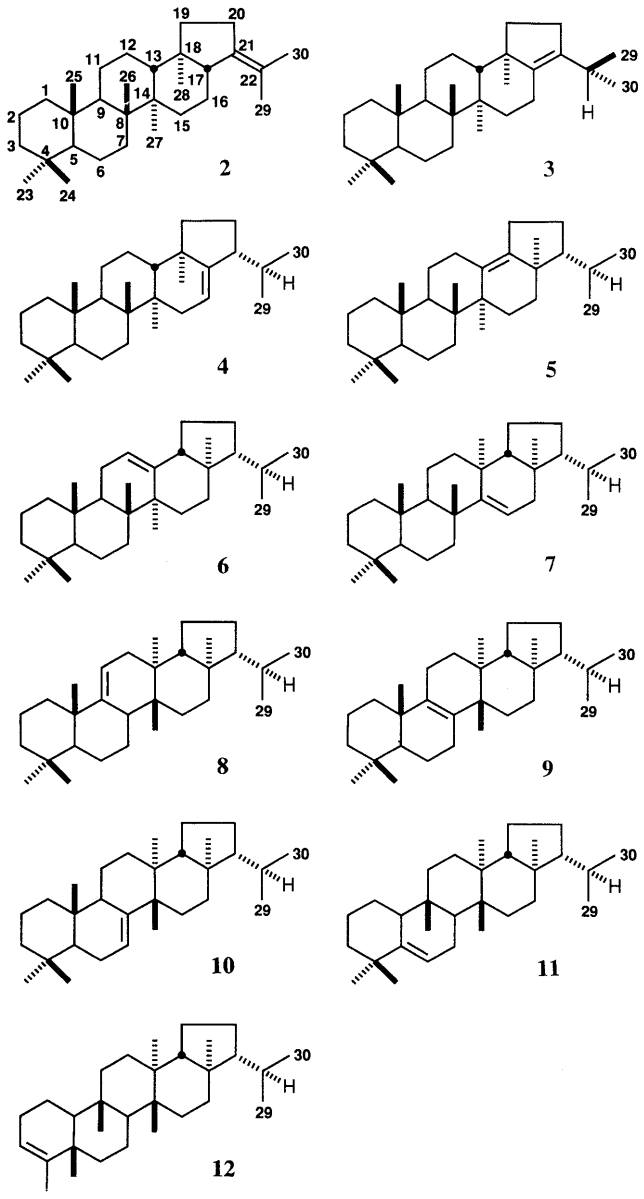

Chart 1

width of 33898.3 Hz were used, giving a digital resolution of 2.1 Hz per point. DEPT and 2D NMR spectra were obtained with the standard JEOL pulse sequences. The H–H COSY and NOESY spectra were obtained at 500.00 MHz. The frequency width was 4450.4 Hz and the initial t_1, t_2 matrix of 512 × 512 real data points was zero-filled to 1024 × 1024 data points to give a final resolution of 4.3 Hz per point. The NOESY spectrum was obtained using a mixing time of 600 ms. The C–H COSY spectrum was obtained using the frequency ranges of 25773.2 and 4449.6 Hz for ^{13}C and ^{1}H, respectively. The initial matrix of 1024 × 256 real data points was zero-filled to 2048 × 512 data points, thus giving digital resolutions of 12.6 and 8.7 Hz per point in the row and column directions, respectively. A sine-bell window function was applied before Fourier transformation; 64 scans were acquired per t_1 increment. In some cases, for sensitivity reasons, the ^{1}H-detected heteronuclear single-quantum coherence (HSQC) spectrum was used with ^{13}C decoupling during acquisition. The HMBC spectrum was recorded at 500.00 MHz with 64 scans (32 dummy scans). The delay τ_1 was set to the value of $1/(2J_{CH})$, 3.6 ms, and τ_2 was set to the value of $1/(2^{2,3}J_{CH})$, 60 ms.

Assignments of NMR Signals and Discussion

The assignments of methyl and olefinic proton signals are listed in Table I, those of methylene and methine proton signals in Table II, and those of all carbon signals in Table III.

Hop-21-ene (2) Although no discrimination was made between H-29 and H-30, or C-29 and C-30, assignments of the methyl signals of this compound were reported from

TABLE I. ^{1}H-Chemical Shifts for Methyl and Olefinic Protons of Triterpenoids belonging to Hopene and Migrated Hopene Series (500 MHz, $CDCl_3$, δ)

Compd. -ene	**2** 21	**3** 17 (21)	**4** 16	**5** 13 (18)	**6** 12	**7** 14	**8** 9 (11)	**9** 8	**10** 7	**11** 5	**12** 3
H-23	0.850	0.851	0.858	0.855	0.862	0.842	0.847	0.876	0.841	1.004	1.575 (dd, 2.0, 3.0)
H-24	0.796	0.792	0.799	0.793	0.815	0.812	0.888	0.828	0.879	1.059	0.975
H-25	0.820	0.825	0.832	0.826	0.880	0.882	1.053	0.942	0.746	0.833	0.892
H-26	0.968	0.931	0.924	0.860	0.732	1.020	0.733	0.951	0.991	0.993	0.912
H-27	0.968	1.042	1.037	1.102	1.121	1.112	0.822	0.770	0.905	0.922	0.921
H-28	0.580	0.837	0.891	0.793	0.757	0.772	0.759	0.770	0.732	0.778	0.775
H-29	1.732	0.977 (d, 6.7)	0.933 (d, 6.2)	0.937 (d, 6.7)	0.938 (d, 6.4)	0.895 (d, 6.7)	0.890 (d, 6.4)	0.888 (d, 6.4)	0.901 (d, 6.7)	0.886 (d, 6.4)	0.886 (d, 6.7)
H-30	1.578	0.917 (d, 6.7)	0.888 (d, 6.2)	0.893 (d, 6.7)	0.847 (d, 6.4)	0.837 (d, 6.7)	0.830 (d, 6.4)	0.827 (d, 6.4)	0.828 (d, 6.7)	0.827 (d, 6.4)	0.825 (d, 6.7)
C=C<u>H</u>–	—	—	5.213 (ddd, 3.7, 3.7, 1.2)	—	5.056 (ddd, 4.2, 2.5, 2.5)	5.285 (dd, 4.2, 3.0)	5.286 (ddd, 5.1, 2.4, 2.4)	—	5.355 (ddd, 3.7, 3.1, 3.1)	5.514 (ddd, 5.8, 2.2, 2.1)	5.161 (br s)
$\Delta(\delta_{H\text{-}29}-\delta_{H\text{-}30})$	0.157	0.060	0.045	0.044	0.091	0.058	0.060	0.061	0.073	0.059	0.061

Signals, unless otherwise stated, are 3H, singlet. Multiplicity and coupling constants (J) are shown in parentheses.

TABLE II. ^{1}H-Chemical Shifts for Methylene and Methine Protons of Triterpenoid Hydrocarbons belonging to Hopene and Migrated Hopene Groups (500 MHz, $CDCl_3$, δ)

Compd. -ene	**2** 21	**3** 17 (21)	**4** 16	**5** 13 (18)	**6** 12	**7** 14	**8** 9 (11)	**9** 8	**10** 7	**11** 5	**12** 3
H-1	0.77; 1.65	0.78; 1.67	0.78; 1.67	0.81; 1.69	0.85; 1.53	0.87; 1.58	1.14; 1.89	1.03; 1.76	0.92; 1.56	1.68; 1.08	1.61; 1.47
H-2	1.37; 1.56	1.40; 1.60	1.38; 1.59	1.40; 1.52	1.38; 1.59	1.35; 1.40	1.44; 1.55	1.43; 1.56	1.45; 1.53	1.60; 1.56	1.99; 2.04
H-3	1.13; 1.35	1.13; 1.34	1.13; 1.34	1.14; 1.34	1.17; 1.37	1.32; 1.35	1.11; 1.36	1.14; 1.37	1.15; 1.41	1.37; 1.19	(5.16)
H-4	—	—	—	—	—	—	—	—	—	—	—
H-5	0.72	0.73	0.73	0.76	0.83	0.85	1.26	1.09	1.35	—	—
H-6	1.49; 1.34	1.48; 1.32	1.50; 1.32	1.54; 1.33	1.57; 1.36	1.55; 1.33	1.72; 1.56	1.68; 1.29	2.16; 1.90	(5.51)	1.24; 1.80
H-7	1.22; 1.45	1.27; 1.45	1.28; 1.55	1.41; 1.60	1.25; 1.40	1.37; 2.00	1.60; 1.33	2.08; 1.87	(5.36)	1.78; 1.83	1.48; 1.36
H-8	—	—	—	—	—	—	2.06	—	—	1.47	1.32
H-9	1.26	1.30	1.24	1.40	1.78	1.69	—	—	2.38	—	—
H-10	—	—	—	—	—	—	—	—	—	2.00	1.24
H-11	1.53; 1.44	1.54; 1.24	1.56; 1.35	1.49; 1.20	2.08; 1.79	1.60; 1.12	(5.29)	1.83; 2.12	1.55; 1.45	1.46; 1.62	1.38; 1.43
H-12	1.46; 1.42	1.38; 1.35	1.58; 1.48	1.90; 2.30	(5.06)	0.90; 0.80	1.62; 1.51	1.27; 1.40	1.33; 1.46	1.55; 1.06	1.48; 1.02
H-13	1.45	1.43	1.44	—	—	—	—	—	—	—	—
H-14	—	—	—	—	—	—	—	—	—	—	—
H-15	1.24; 1.36	1.26; 1.26	1.56; 2.16	1.28; 1.87	1.67; 1.00	(5.29)	1.40; 1.32	1.33; 1.72	1.59; 1.48	1.38; 1.18	1.37; 1.22
H-16	1.61; 2.16	1.90; 2.26	(5.21)	1.79; 1.24	1.93; 1.65	2.18; 1.99	1.65; 1.40	1.65; 1.46	1.73; 1.60	1.58; 1.60	1.58; 1.67
H-17	1.72	—	—	—	—	—	—	—	—	—	—
H-18	—	—	—	—	2.12	1.35	1.56	1.53	1.48	1.59	1.57
H-19	1.52; 1.02	1.64; 1.31	1.69; 1.02	2.20; 2.27	1.30; 1.65	1.28; 1.51	1.35; 1.35	1.28; 1.40	1.29; 1.38	1.25; 1.35	1.20; 1.37
H-20	2.19; 2.07	2.18; 2.11	1.43; 1.82	1.37; 1.83	1.22; 1.87	1.28; 1.84	1.21; 1.83	1.21; 1.82	1.21; 1.82	1.20; 1.82	1.21; 1.82
H-21	—	—	2.04	1.03	1.27	1.01	0.97	0.97	0.95	0.98	1.00
H-22	—	2.64	1.50	1.55	1.48	1.45	1.45	1.46	1.45	1.44	1.45

Methylene signals are listed as α-H; β-H in each column; assignments were confirmed by NOESY spectra, splitting patterns, and a consideration of the relation to the double bond.

TABLE III. ^{13}C-Chemical Shifts for Triterpenoids belonging to Hopene and Migrated Hopene Series (125 MHz, $CDCl_3$, δ)

Compd. -ene	**2** 21	**3** 17 (21)	**4** 16	**5** 13 (18)	**6** 12	**7** 14	**8** 9 (11)	**9** 8	**10** 7	**11** 5	**12** 3
C-1	40.32	40.44	40.31	40.57	39.63	39.88	41.49	37.28	38.63	25.89	17.50
C-2	18.72	18.72	18.70	18.75	18.41	18.61	19.56	19.28	19.08	21.82	27.22
C-3	42.12	42.13	42.10	42.07	42.16	42.13	42.43	41.80	42.41	40.82	120.30
C-4	33.27	33.30	33.26	33.31	33.11	33.18	33.64	33.35	33.17	35.70	144.33
C-5	56.14	56.31	56.23	56.79	56.58	57.05	44.88	51.11	51.48	145.54	38.44
C-6	18.72	18.72	18.47	18.85	18.92	20.68	19.53	19.41	24.58	117.55	38.73
C-7	33.22	33.45	33.49	34.47	32.61	43.14	17.90	18.90	116.21	23.76	18.27
C-8	41.87[a]	41.93[a]	41.42[a]	41.44	41.69	39.09	39.98	133.68	145.35	44.23	49.36
C-9	50.41	50.93	50.42	52.28	48.12	48.83	151.68	134.90	48.00	34.71	37.62
C10	37.41	37.49	37.38	37.77	37.89	37.85	38.05	37.74	35.53	51.67	57.16
C-11	20.93	21.29	20.89	21.55	23.60	33.40	115.60	26.93	16.07	34.26	35.20
C-12	23.71	24.10	24.18	26.75	118.18	16.07	36.78	30.32	32.41	29.10	28.47
C-13	48.07	49.32	44.80	131.71	145.37	36.18	36.74	36.70	36.03	38.60	39.06
C-14	41.47[a]	42.03[a]	40.50[a]	42.38	39.46	156.88	37.69	41.05	41.57	39.35	40.22
C-15	32.81	31.82	34.04	29.28	25.01	117.64	29.28	26.93	30.29	29.12	29.13
C-16	23.29	19.86	119.63	37.95	34.65	43.45	36.19	35.96	36.29	35.45	35.66
C-17	56.00	140.07	147.93	42.67	40.07	40.72	42.97	42.90	42.86	42.81	42.77
C-18	44.36	49.85	43.63	141.10	52.64	58.90	52.02	52.76	54.16	51.79	51.76
C-19	39.09	41.65	43.22	26.50	22.84	19.58	20.15	20.37	20.00	19.93	19.93
C-20	28.41	27.51	27.10	27.57	28.41	28.41	28.23	28.38	28.25	28.34	28.42
C-21	135.61	135.99	51.39	59.17	60.10	59.45	59.68	59.78	59.57	60.06	60.09
C-22	120.58	26.37	35.07	29.81	31.73	30.78	30.80	30.76	30.68	30.80	30.77
C-23	33.43	33.41	33.45	33.31	33.43	33.36	32.80	33.26	32.92	29.93	17.94
C-24	21.61	21.60	21.65	21.55	21.57	21.55	21.68	21.75	21.19	29.73	20.65
C-25	15.87	16.20	15.97	16.73	15.22	15.97	25.06	20.25	12.82	17.92	20.61
C-26	16.63[b]	16.37	16.93	18.72	16.39	27.98	15.84	22.13	24.05	15.75	16.08
C-27	16.61[b]	14.99	18.03	26.70	22.28	20.78	15.43	15.76	21.06	15.01	15.65
C-28	14.68	19.10	19.31	17.87	18.57	16.04	14.00	14.63	14.03	16.07	16.34
C-29	19.40	21.93	22.06	22.88	22.51	21.98	22.14	22.06	22.11	21.97	21.95
C-30	22.81	21.32	21.53	23.08	22.66	22.95	23.02	22.98	23.00	22.93	22.91
$\Delta(\delta_{C\text{-}29}-\delta_{C\text{-}30})$	−3.41	+0.61	+0.53	−0.20	−0.15	−0.97	−0.88	−0.92	−0.89	−0.96	−0.96

a, b) Assignments of the signals might be reversed.

our laboratory,[2,3,9] and the carbon signals were assigned by Wilkins *et al.*[12] As in hopane derivatives,[1] precise assignments for C-8 and C-14 of compounds **2**, **3** and **4** are very difficult because the HMBC method is not applicable. In addition, the chemical shifts for C-2 and C-6 of compounds **2** and **3** showed the same value, and thus assignments of H-2 and H-6 were difficult. Carbon signals for C-1, 3, 4, 5, 7, 9, 10, 13, 15, 17, 18, 19, 21 and 22 were assigned from the HMBC spectrum with the H-23, 24, 25, 26, 27, 28, 29 and 30 signals without ambiguity. Other carbon signals (C-2, 6, 11, 12, 16 and 20) were assigned from the corresponding proton signals, confirmed by the H–H-COSY spectrum with the signals of proton(s) attached to the neighboring carbon(s). The conformation of this compound with minimum steric energy (85.843 kcal/mol) was simulated as shown in Fig. 1, and all cross peaks observed in the NOESY spectrum except the peaks between two protons attached to the same carbon are shown in Fig. 1. The NOE peaks needed to establish the assignments of H-29 and H-30 were observed between H-29 and H-16β (δ 2.16), H-16β and H-17; H-30 and H-20α (δ 2.19), H-20α and H-28; H-30 and H-20β (δ 2.07). The side chain conformation was found to be a new type (type F), different from the previous types A—E.[1] Assignments of all proton signals of **2** are shown in Tables I and II, and those of the corresponding carbon signals in Table III. The proton signals of H-16β, H-17, H-20α and H-20β were observed at lower fields, and H-28 at higher field owing to the anisotropic effect of the double bond.

Hop-17(21)-ene (3) Assignments of ^{1}H-NMR signals of this compound were published by us,[2,3,5,9] and those of ^{13}C-NMR by Wilkins *et al.*[12] and by us,[3] without evidence to confirm the assignments of H-29 and H-30, or C-29 and C-30 signals. Most of the proton and carbon signals were assigned as shown in Tables I—III. The conformation with minimum steric energy (75.787 kcal/mol) of this compound was simulated as shown in Fig. 1, and this was supported by the NOESY spectrum. Cross peaks between H-22 and H-16β (δ 2.26), H-29 and H-20β (δ 2.11), H-30 and H-20α (δ 2.18), and H-28 and H-16α (δ 1.90), H-20α (δ 2.18), clearly indicated that the side chain conformation of **3** was similar to that of isohopane (type C).[1] Thus, the assignments of H-29 and H-30, and C-29 and C-30 were established. The proton signals of H-16, H-20, H-22, H-28, H-29 and H-30 were observed at lower fields owing to the anisotropic effect of the double bond.

Hop-16-ene (4) This compound is a rare natural product only obtained so far from the rhizomes of *Davallia mariesii*, and the ^{1}H- and ^{13}C-NMR data were reported by us.[3] The partly revised assignments of the proton and carbon signals were established by the procedure in this paper (Tables I—III). The conformation of this compound

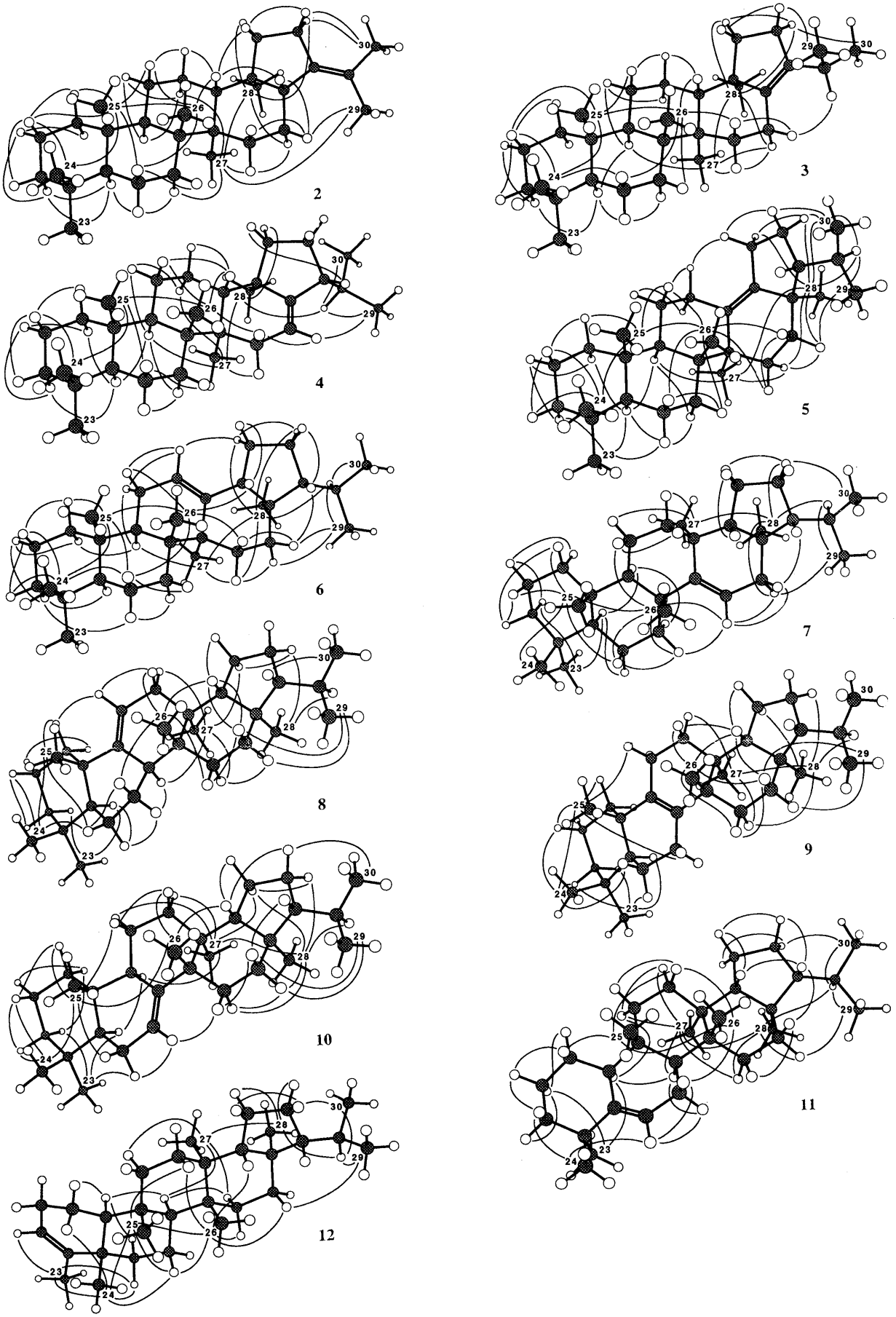

Fig. 1. Chem3D Plus Drawing and NOEs (—)

with minimum steric energy (86.267 kcal/mol) was simulated as shown in Fig. 1, and was supported by the NOESY spectrum. The side chain part was of the hopane type (type A),[1] as revealed by cross peaks in the NOESY spectrum between H-22 and H-28; H-22 and H-29, H-29 and H-16, H-16 and H-15β (δ 2.16). The signals of H-15β, H-20β, H-21, H-28, H-29 and H-30 were found to be shifted remarkably to lower fields, and H-19β to higher field by owing to the anisotropic effect of the double bond.

Neohop-13(18)-ene (5) The assignments for the methyl protons of this compound were reported by us.[2,4,9] The signals of methylene and methine protons were assigned as shown in Table II. Thirty carbon signals were assigned without ambiguity except C-8 and C-14. The conformation of this compound with minimum steric energy (75.781 kcal/mol) was simulated as shown in Fig. 1, and was supported by the NOESY spectrum. The side chain part was confirmed to be of the hopane type (type A)[1] by cross peaks in the NOESY spectrum between H-22 and H-28, H-28 and H-27; H-29 and H-16α (δ 1.79), H-16α and H-28; H-30 and H-20α (δ 1.37), H-20α and H-28. Thus, the signals of H-29 and H-30 as well as C-29 and C-30 were firmly assigned. The conformations of rings C (a chair form) and D (a half boat form) were supported by the fact that proton signals of H-12, H-19, H-27 and H-28 were observed at lower fields and H-16β at higher field owing to the anisotropic effect of the double bond.

Neohop-12-ene (6) The correct assignments for the methyl protons except H-29 and H-30 have been reported from our laboratory.[4,5,9] All carbon signals were readily assigned by the procedure without ambiguity (Table III). Comparing the conformation with minimum steric energy (80.123 kcal/mol) simulated by Chem3D Plus and MM2 (Fig. 1) with a Dreiding model, the ring D (boat form with two methyls at the bows) conformation of the former was different from that of the latter because of interaction between C-27 and C-28 methyls. This is the reason why **6** is rather unstable,[10] and easily transformed to **5**. The conformation of the side chain (the hopane-type) was proved by cross peaks in the NOESY spectrum between H-22 and H-28, H-28 and H-27; H-29 and H-16α (δ 1.93), H-16α and H-15α (δ 1.67), H-15α and H-27; H-29 and H-16β (δ 1.65), H-16β and H-15β (δ 1.00), H-15β and H-26. Thus H-29 and H-30, as well as C-29 and C-30, were definitely assigned. The methylene and methine protons were assigned as shown in Table II. The proton signals of H-11α, H-15α, H-18 and H-27 were shifted to lower fields, and H-15β to higher field, owing to the anisotropic effect of the double bond.

Pteron-14-ene (7) Although this compound was synthesized from fern-7-ene (**10**) and supposed to be a stable member of the migrated hopanes, no one has yet been able to isolate this compound or its derivative from a natural source. The methyl signals were assigned by us,[5] but the values for H-23 and H-25 were erroneously reported. The carbon signals except those of C-29 and C-30 were readily assigned as shown in Table III without ambiguity. The stable conformation of the compound with minimum steric energy (78.375 kcal/mol) was simulated, and was supported by cross peaks in the NOESY spectrum as shown in Fig. 1. The conformation of ring C was found to be a boat form, and that of the side chain was proved to be of the hopane-type by NOEs between H-22 and H-28, H-29 and H-16α (δ 2.18), H-30 and H-20α (δ 1.28). The signals of H-7β (δ 2.00), H-16α, H-16β (δ 1.99), H-26 and H-27 were observed at lower fields owing to the anisotropic effects of the double bond.

Fern-9(11)-ene (8) Assignments for the methyl protons based on $CDCl_3$–C_6D_6 solvent shifts reported by us[5,9] were found to be correct except those for H-29 and H-30. ^{13}C-NMR assignments were reported by Wilkins *et al.*,[12] and also by us,[13] but the signals for C-2 and C-6, and C-29 and C-30 were not discriminated. The conformation of the compound with minimum steric energy (74.291 kcal/mol) was simulated, and was supported by cross peaks in the NOESY spectrum as shown in Fig. 1. The signals of all methylene and methine protons were assigned as shown in Table II, and those of C-2 and C-6 were established by the H–H-COSY spectrum of the corresponding proton signals. The conformation of ring B was a boat form, and that of ring C a twist form. The side chain conformation was proved to be of the hopane-type (type A)[1] by NOEs between H-22 and H-28; H-29 and H-16α (δ 1.65) or H-16β (δ 1.40). Thus, the signals of H-29 and H-30, as well as C-29 and C-30 were assigned as shown in Tables I and III. The signals of H-1β, H-6α, H-7α, H-8, H-12α, H-25 were remarkably shifted to lower fields, and H-26 to higher field, owing to the anisotropic effect of the double bond.

Fern-8-ene (9) Assignments for the methyl protons reported by us[2,9] were found to be correct except those for H-29 and H-30. Most of the carbon signals were assigned by the HMBC, and those of C-2, 6, 11, 19 and 20 by the H–H-COSY of the corresponding proton signals as shown in Table III, without ambiguity. The stable conformation with minimum steric energy (73.338 kcal/mol) was simulated as shown in Fig. 1, and was supported by cross peaks in the NOESY spectrum. The conformations of the rings C, D and E were conversed compared with those of hopane, and rings B and C were both twist forms. All methylene and methine protons were assigned as shown in Table II. The conformation of the side chain was found to be of the hopane-type (type A),[1] which was proved by NOEs between H-22 and H-28; H-29 and H-16α (δ 1.65) or H-16β (δ 1.46). Thus, the signals of H-29 and H-30, as well as C-29 and C-30, were firmly assigned. The proton signals of H-1β, H-6α, H-7α, H-11β and H-15β were observed at lower field, and H-27 at higher field, owing to the effect of the double bond.

Fern-7-ene (10) Assignments for the methyl protons reported from our laboratory[2,5,9,14,15] were found to be correct except those for H-29 and H-30. Most of the carbon signals were readily assigned as shown in Table III. The stable conformation with minimum steric energy (78.052 kcal/mol) was simulated and supported by cross peaks in the NOESY spectrum as shown in Fig. 1. The conformations of rings D and E were conversed compared to those of hopane, ring B was found to be in a half boat form, and the ring C a twist form, while the side chain was of the hopane-type. The signals of methylene and methine protons were assigned as shown in Table II. Finally, the signals of H-29 and H-30, and C-29 and C-30

were assigned as shown in the tables. Owing to the anisotropic effect of the double bond, the proton signals of H-6, H-9 and H-26 were shifted to lower fields, and those of H-25 and H-27 to higher fields.

Adian-5-ene (11) Assignments for the methyl groups based on $CDCl_3$–C_6D_6 solvent shift[9] were found to be correct except those of H-29 and H-30. The carbon signals were readily assigned except those of C-29 and C-30, and C-12 and C-15, the signals of the latter two being very close (δ 29.10 and 29.12). The conformation of **11** with minimum steric energy (79.217 kcal/mol) was simulated as shown in Fig. 1, and was supported by cross peaks in the NOESY spectrum. It is interesting that the conformations of rings A, B, C, D and E are completely conversed compared with those of hopane.[1] The signals of methylene and methine protons were assigned as shown in Table II, and the conformation of the side chain was proved to be of hopane type by cross peaks in the NOESY spectrum between H-22 and H-28, H-29 and H-16α (δ 1.58), H-16α and H-28; H-29 and H-16β (δ 1.60); H-30 and H-20β (δ 1.82), H-20β and H-21. Thus, the signals of H-29 and H-30, and C-29 and C-30 were completely assigned. The anisotropic effect of the double bond shifted the proton signals of H-1, H-7, H-10, H-23 and H-24 to lower fields, and that of H-25 to higher field.

Filic-3-ene (12) Assignments for the methyl signals based on $CDCl_3$–C_6D_6 solvent shift were reported by us,[3,16] and those for the carbon signals[16] were found to be correct by the procedure of this paper (Tables I and III) except H-29, H-30 and C-29, C-30. The conformation of **12** with minimum steric energy (86.613 kcal/mol) was simulated as shown in Fig. 1, being conversed in all rings like **11**. The facts that 1) the proton, H-3, attached to the double bond gave a broad singlet signal and no splitting pattern was observed; 2) no cross peaks in the NOESY spectrum were observed between H-2α or H-2β and other protons; and 3) a very high value of steric energy was obtained, may suggest that the conformation of ring A of **12** is variable at C-2. The signals of methylene and methine protons were assigned as shown in Table II. The hopane-type conformation of the side chain was supported by the cross peaks in the NOESY spectrum between H-22 and H-28; H-29 and H-16β (δ 1.67); H-30 and H-20β (δ 1.82). Thus, the signals of H-29 and H-30, as well as C-29 and C-30 were firmly assigned without ambiguity. The proton signals of H-1, H-2, H-6β and H-24 were observed at lower field owing to the effect of the double bond.

Additional Discussion for the Conformations

The conformations of eleven compounds simulated by Chem3D Plus and MM2 were supported by NOESY spectra in the solution. Migration of the double bond from ring E to ring A, causes the right-hand side of the rings to take the converse conformation compared with those of hopane. The steric energies of the compounds are in the order **9*** < **8** < **5*** < **3*** < **10** < **7** < **11** < **6** < **1** < **2*** < **4** < **12**, in which the compounds marked with * have a tetrasubstituted double bond and the others have a trisubstituted one. Compounds **9**, **5** and **3** are the stable products of acid rearrangements,[10] while compound **8** is the most widely distributed hydrocarbon among fern plants. As far as the conformation of the side chains is concerned, that of compound **2** was a new type (type F) and that of **3** was type C,[1] while those of compounds **4**—**12** were found to be hopane type (type A).[1] For comparison of the side chain signals, $\Delta(\delta_{H\text{-}29}-\delta_{H\text{-}30})$ and $\Delta(\delta_{C\text{-}29}-\delta_{C\text{-}30})$ of compounds **2**—**12** are shown in Tables I and III. $\delta_{H\text{-}29}$ is observed at lower field than $\delta_{H\text{-}30}$ in every case, just as in hopane,[1] while $\delta_{C\text{-}29}$ appears at higher field than $\delta_{C\text{-}30}$ except in **3** and **4**.

References and Notes

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