TRITERPENOIDS FROM THE LEAVES OF FAR EASTERN SPECIES OF THE SHRUBBY BIRCHES *Betula ovalifolia* AND *B. middendorfii**

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From the unsaponifiable fraction of an ethereal extract of the leaves of *Betula ovalifolia* have been isolated the new triterpene 20(S),24(R)-epoxydammarane-3α,17α,25-triol (V) and the corresponding monoketone at C^3 (VI). The leaves of *B. middendorfii* have yielded the triterpene (IX) and (X), identified as, respectively, dammar-23-ene-3α,12β,20(S),25-tetraol and damman-25-ene-3α,12β,20-(S),24-tetraol, which have been obtained previously from the leaves of *Betula platyphylla* Sukatchev var. *japonica*.

In the study of the triterpenoid composition of the leaves of *Betula ovalifolia*, together with the known 3-epiocotillol (I), betulafolienetriol oxide (II), betulafolienetetraol oxide (III), and 20(S),24-dihydroxydammar-25-en-3-one (IV) [1-5] we have detected two new triterpenes (V) and (VI).

The IR spectrum of the triterpene (V) in $CHCl_3$ solution (c 35.0 mg/ml) shows bands at 3433, 3446, 3578, and 3608 cm^{-1}, characteristic for the stretching vibrations of a hydroxy group. On 15-fold dilution of the solution, the band at 3433 cm^{-1} disappeared, and the bands at 3446 and 3578 cm^{-1} remained practically unchanged.

In the ^1H spectrum of (V), the signals of the protons of eight tertiary methyl groups appear in the strong field at 0.83-1.23 ppm, and the signal of a carbinyl proton in the weak field at 3.37 ppm (1 H, triplet, J = 2.8 Hz), which for the triterpenes of the dammarene series can be unambiguously assigned to H^3. The values of δ and of J for the H^3 signal show the α configuration of the hydroxy group at C^3 [5, 6]. A signal also appears in the weak field at 3.76 ppm (1 H, triplet, J = 6 Hz), which can be assigned to H^{24}, since it is observed in the ^1H spectra of all dammarane triterpenes with a side chain of ocotillone type [5, 6].

The presence in the mass spectrum of a triterpene (V) peak with m/e 143 (100%) indicates that the side chain of triterpene (V) is a substituted tetrahydrofluorane ring [6].

The ^{13}C spectrum of (V) (Table 1) contains the signals of five carbinyl C atoms at 76.3 (d), 84.2 (s), 83.7 (d), 72.0 (s), and 90.3 (s) ppm. By comparing the ^{13}C spectra of 3-epiocotillol (I) and of betulafolienetetraol oxide (III) with the ^{13}C spectrum of the triterpene (V) it was found that (V) is the 17-hydroxy analog of (I) and the 12-deoxy analog of (III), while the effects of the 17-OH group in a comparison with the ^{13}C of (I) and (V) show its α configuration [7].

The chemical shifts (CSs) of the carbinyl C atoms mentioned above therefore relate to the signals of the C^3, C^{17}, C^{24}, C^{25}, and C^{20} atoms, respectively. The good agreement of the CSs of the C^{21}, C^{22}, C^{23}, C^{24}, and C^{25} atoms with the ^{13}C spectra of 20(S),24(R)-3-epiocotillol (I), 20(S),24(R)-epoxydammarane-3α,12β,17α,25-tetraol (III), and the triterpene (V) show a correspondence of the stereochemistry of the C^{20} and C^{24} asymmetric centers in these compounds. Acetylation of the triterpene (V) under severe conditions yielded a monoacetate at C^3 (VII) and a diacetate at C^3 and C^{25} (VIII). The IR spectrum of (VIII) in $CHCl_3$ solution (c 50.5 mg/ml) showed hydroxyl absorption at 3415, 3573, and 3603 cm^{-1}. With a 23-fold dilu-

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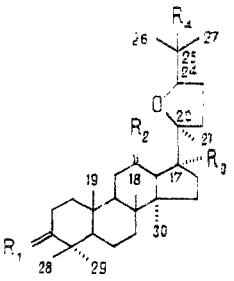


I $R_1 = \alpha\text{-OH}$, H; $R_2 = H_2$; $R_3 = H$; $R_4 = OH$

II $R_1 = \alpha\text{-OH}$, H; $R_2 = \alpha\text{-OH}$, H; $R_3 = H$; $R_4 = OH$

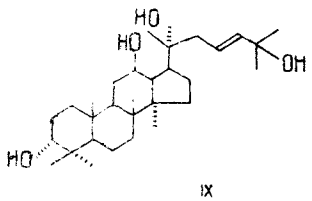
III $R_1 = \alpha\text{-OH}$, H; $R_2 = \alpha\text{-OH}$, H; $R_3 = OH$; $R_4 = OH$

V $R_1 = \alpha\text{-OH}$, H; $R_2 = H_2$; $R_3 = OH$; $R_4 = OH$

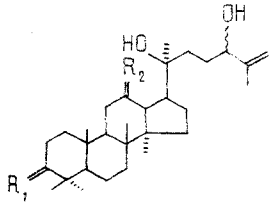
VI $R_1 = O$; $R_2 = H_2$; $R_3 = OH$; $R_4 = OH$

VII $R_1 = \alpha\text{-}OCOCH_3$ (31, 32), H; $R_2 = H_2$; $R_3 = OH$; $R_4 = OH$

VIII $R_1 = \alpha\text{-}OCOCH_3$ (31, 32), H; $R_2 = H_2$; $R_3 = OH$; $R_4 = OCOCH_3$ (33, 34)

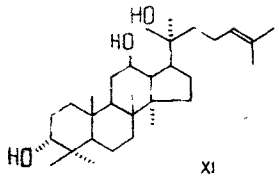


IX



IV $R_1 = O$; $R_2 = H_2$

X $R_1 = \alpha\text{-OH}$, H; $R_2 = \alpha\text{-OH}$, H



XI

tion of the solution, the band at 3415 disappeared and the band at 3573 cm^{-1} did not change its position or intensity. On the basis of these facts, the structure of 20(S),24(R)-epoxydammarane-3α,17α,25-triol is proposed for the triterpene (V).

The IR spectrum of the triterpene (VI) in $CHCl_3$ solution (c 38 mg/ml) shows the band of the stretching vibrations of a carbonyl group in a six-membered ring at 1695 cm^{-1} and bands characteristic for the stretching vibrations of a hydroxy group at 3432, 3441, 3575, and 3601 cm^{-1}. With a 20-fold dilution of the solution, the band at 3432 cm^{-1} disappeared, and the bands at 3411 and 3575 cm^{-1} underwent practically no change.

By the ^1H spectrum of (VI) the signals of the protons of eight tertiary methyl groups appear in the strong field at 0.94-1.24 ppm, and the signal of a carbinyl proton in the weak field at 3.77 ppm (1 H, triplet, J = 6.7 Hz) corresponding to H^{24}.

The ^{13}C spectrum of (VI) contains the signal of a carbonyl C atom, which can be unambiguously assigned to C^3 [4, 5]. The good agreement of the CSs of the C atoms in the ^{13}C spectra of the triterpenes (V-VIII) after allowing for the influence of the carbonyl and acetyl groups [7, 8], permits us to speak of the identity of their skeletons.

When the triterpene (V) was oxidized with CrO_3 in pyridine, the monoketone at C^3 was obtained, which was identical in relation to melting point and spectral characteristics with

TABLE 1. ^{13}C Chemical Shifts of Compound (I), (III), and (V-XI) (ppm relative to TMS, $CdCl_3$)

C atom	Compound								
	I	III	V	VI	VII	VIII	IX	X	XI
1	33.7	33,6	33,8	40,0	34,6	34,5	33 8	33,8	33,8
2	25.4	25,5	25,5	34,0	23,0	23,0	25,5	25 5	25,3
3	76.1	76,2	76.3	217,7	78 5	78,7	76,1	76,1	76,0
4	37,6	37,4	37,5	47.4	36,8	36,9	37,7	37.7	37 6
5	49,5	49,6	49,6	5,4	50,8	50,9	49,5	49,6	49,5
6	18,2	18,3	18,4	19,8	18.4	18,3	18,4	18,4	18,1
7	35,4	34,2	34,8	34,0	34,8	34,8	34,9	34,9	34,8
8	40,6	40,6	41,3	40,9	41,4	41,3	40,0	40,1	39,9
9	50 6	50,5	50.8	50,3	50 9	50,3	50,0	50,1	50,0
10	37,3	37,6	37.7	37,1	37,3	37,4	37.4	37.4	37,2
11	21,4	31,7	21,6	22.2	22,2	21,6	31,1	31,2	31,3
12	27,4	67,8	23,3	23,4	23,3	23,2	71,0	71,1	70,8
13	42,9	52,0	45,6	45,9	45,8	45,7	48,2	47.7	47,5
14	50,2	51,3	50,0	49.8	50,0	50,0	51,7	51,9	51,7
15	31,5	33.7	32,7	32,6	32.7	32,7	31,1	31,2	31,0
16	25,7	38,7	37,2	37,1	37,2	37,1	26,3	26,6	26 5
17	49,5	85,6	84,2	83,9	84,1	83,6	53,0	53,8	53,6
18	16,1	16,3	16,2	16,1	16,2	16,2	16,1	16,1	16,0
19	15,5	15,8	15,8	15,3	15,8	15,8	15,9	15.7	15,7
20	86,4	80,8	90,3	90,1	90,2	90,3	73,6	73,4	73,8
21	23,6	22,1	22,3	22,2	22,3	22,3	27,0	26,6	26,7
22	35,7	33,6	33,1	33,1	33,0	32,7	38,1	30,2	34,8
23	26,2	25,2	26,4	26,5	26,7	26,6	122,6	29,8	22,4
24	83,3	85,7	83,7	83,6	83,7	83,1	141,7	75,4	125,3
25	71,4	70,1	72,0	71.9	72,1	83,1	70,3	147,5	131,2
26	27,4	28,1	27,3	27.5	27 2	22,7	30,0	110.5	25,8
27	24,3	26,3	25,2	25,2	25,3	22,3	28,6	18,4	17.7
28	28,4	28,3	28,4	26,8	27,8	27,9	28,6	28,5	28,4
29	22,1	22,1	22,3	21,0	21,7	21,8	22,3	22,2	22,2
30	16,6	18,5	17,3	17,1	17,3	17,4	17,2	17,0	17,0
31					170,5	170,7			
32					21,2	21,3			
33						171,1			
34						21,6			

the triterpene (VI). Thus, for triterpene (VI) the structure of 17α,25-dihydroxy-20(S),24-(R)-epoxydammaran-3-one is proposed.

We have isolated four triterpenes from the leaves of the Far Eastern species *Betula middendorffii* — betulafolienetriol (XI) and its oxide [1-3] and also the triterpenes (IX) and (X).

On the basis of the comparison of the ^1H and ^{13}C spectra of the triterpenes (IX), (X), and (XI), compounds (IX) and (X) have been assigned the structures dammar-23-ene-3α,12β,20-(S),25-tetraol and dammar-25-ene-3α,12β,20(C),24-tetraol, respectively. From the results of PMR and IR spectroscopy and mass spectrometry, compounds (IX) and (V) proved to be identical with triterpenes isolated previously from the leaves of *Betula platyphylla* Sukatchev var. *japonica* [9].

In view of the determination of the structures of triterpenes (V) and (VI) particular interest is presented by the results given above of an investigation of their solutions in $CHCl_3$ by IR spectroscopy. Bands at 3446 and 3578 cm^{-1} in the IR spectrum of (V), like those at 3441 and 3575 cm^{-1} in the IR spectrum of (VI), are assigned to the stretching vibrations of hydroxy groups bound by intramolecular hydrogen bonds (intra-HBs), the peak intensity and integral intensity of the low-frequency band being higher than those of the high-frequency band in both cases. Two variants of an intra-HB are possible for the molecules of (V) and (VI): through the interaction of the proton of the C^{25}—OH group with the oxygen atom of the tetrahydrofuran (THF) ring, and of the proton of the C^{17}—OH group with the oxygen atom of the C^{25}—OH group, or conversely. In the IR spectrum of the triterpene (VIII) [the acetyl derivative of the triterpene (V) at the C^{25}—OH group] only one intra-HB band is observed, at 3573 cm^{-1}, which apparently relates to a bond between the proton of the C^{17}—OH group and the oxygen atom of the THF ring. At the same time, in the IR spectrum of 3-epiocotillol (I) the band of the intra-HB assigned to the interaction of the proton of the C^{25}—OH group with the oxygen atom of the THF ring is observed at 3560 cm^{-1}. A similar weak intra-HB (3580 cm^{-1}) has been reported previously for ocotillol [10].

A comparison of the IR spectra of triterpenes (I) and (VIII) with the spectra of (V) and (VI) shows that one of the intra-HBs in the molecules of (V) and (VI) is analogous to the intra-HB in (VIII), i.e., it relates to the interaction of the proton of the C^{17}—OH group with the oxygen atom of the THF ring. The second intra-HB in the molecules of (V) and (VI) consequently relates to the interaction of the proton of the C^{28}—OH group with the oxygen atom of the C^{17}—OH group, and, in spite of the large size of the ring formed through this bond, its energy is higher than that of the first intra-HB, which is probably due to the influence of the first intra-HB.

EXPERIMENTAL

IR spectra were recorded on a Specord 75 IR spectrophotometer in $CHCl_3$ solution. Mass spectra were obtained on a LKB 9000 spectrometer at 70 eV. ^1H and ^{13}C NMR spectra were measured on a Bruker HX-90 E instrument for solutions of the triterpenes in $CHCl_3$, with TMS as internal standard. Chemical shifts (CSs) are expressed in the δ scale. The accuracy of the measurement was ±0.15 Hz for ^1H and ±1.5 Hz for ^{13}C. The assignment of the signal in the ^{13}C spectra of the triterpenes was performed by the method of off-resonance spin decoupling and by selective double heteronuclear resonance on the basis of the PMR characteristics.

The individuality of the substances was checked by TLC on silica gel (KSK) in the chloroform—ethanol (10:1), petroleum ether—acetone (2:1), and benzene—ethanol (1:1) systems. A 10% solution of H_2SO_4 in methanol was used to detect the triterpenes on the chromotograms. Triterpenes (I-IV and XI) were identified by comparison with authentic samples with respect to melting points and IR spectra. The elementary analyses of all the compounds corresponded to the calculated figures.

Isolation of the Triterpenes (I-VI). The air-dry leaves of *B. ovalifolia* (5 kg), collected in July 1979 in the village of Novoselishche, Khanka region of the Maritime Territory were exhaustively extracted with diethyl ether at room temperature. The extract was evaporated and was treated by the method of Fischer and Seiler [11]. The unsaponifiable fraction of the ethereal extract (28 g) was chromatographed on a column of SiO_2 with elution by the hexane—acetone (40 → 1) system. Subsequent repeated recrystallization of the residues from the evaporation of the eluates gave the individual triterpenes (I-VI).

Triterpene (V). $C_{30}H_{52}O_4$, 540 mg, mp 191-193°C (hexane—acetone), $[\alpha]_D^{20}$ +9.0° (c 0.5; $CHCl_3$).

^1H spectrum (ppm); 0.83 (s, 3H); 0.86 (s, 3H), 0.94 (s, 3 H) 0.96, (s, 3 H), 1.13 (s, 3 H), 1.16 (s, 3 H), 1.21 (s, 3 H), 1.23 (s, 3 H), 3.37 (1 H, t, J = 2.8 Hz, H_e^3), 3.76 (1 H, t, J = 6.6 Hz, H^{24}).

Mass spectrum (m/e): 476, 458 ($M^+ - H_2O$), 443 ($M^+ - H_2O - CH_3$), 425 ($M^+ - CH_3 - 2H_2O$), 399 ($M^+ - 59 - H_2O$), 333 ($M^+ - 143$), 315 ($333 - H_2O$), 143 (100%), 125, 59.

Triterpene (VI). $C_{30}H_{50}O_4$, 40 mg, mp 179-182° (hexane—acetone), $[\alpha]_D^{20}$ +58.4° (c 0.5; $CHCl_3$).

^1H spectrum (ppm): 0.94 (s, 3 H), 0.99 (s, 3 H), 1.04 (s, 3 H), 1.08 (s, 3 H), 1.14 (s, 3 H), 1.15 (s, 3 H), 1.22 (s, 3 H), 1.24 (s, 3 H), 3.77 (1 H, t, J = 6.7 Hz, H^{24}).

Mass spectrum (m/e): 474, 456 ($M^+ - H_2O$), 441 ($M^+ - H_2O - CH_3$), 397 ($M^+ - 59 - H_2O$), 331 ($M^+ - 143$), 313 ($331 - H_2O$), 143 (100%), 125, 59.

Oxidation of the Triterpene (V). A solution of 40 mg of the triterpene (V) in 1 ml of absolute pyridine was added dropwise to a solution of 90 mg of CrO_3 in 1.5 ml of pyridine. The reaction mixture was left overnight at room temperature. After the usual working up, the residue was recrystallized twice from a mixture of hexane and acetone. This gave 14 mg of the monoketone at C^3 with mp 178-181°C, which showed no depression of the melting point in a mixture with the triterpene (VI).

Acetylation of the Triterpene (V). A solution of 83 mg of the triterpene (V) in 1 ml of pyridine was treated with 1.4 ml of acetic anhydride. The reaction mixture was heated at 90°C for 24 h and was then cooled to 25°C and poured onto ice. The precipitate was filtered off, washed with water, dried, and separated on a column of SiO_2. On elution with hexane—acetone (60:1), 60 mg of the noncrystalline diacetate (VI) was obtained. Elution with the hexane—acetone (30:1) system gave 20 mg of the noncrystalline monoacetate (VII).

The Monoacetate at C_3 (VII). $C_{32}H_{54}O_5$. $[\alpha]_D^{20}$ −6.8 (c, 0.5; $CHCl_3$). IR spectrum, (ν, cm^{-1}): 1714, 3423, 3475, 3583, 3603.

^1H spectrum (ppm): 0.84 (s, 3 H), 0.88 (s, 6 H), 0.97 (s, 3 H), 1.13 (s, 3 H), 1.19 (s, 3 H), 1.22 (s, 3 H), 1.24 (s, 3 H), 2.09 (s, 3 H, $OCOCH_3$), 3.76 (1 H, t, J = 6.8 Hz, H^{24}), 4.62 (1 H, t, J = 2.7 Hz, H_e^3).

The Diacetate at C^3 and C^{25} (VIII). $C_{34}H_{56}O_6$. $[\alpha]_D^{20}$ −15.8 (c, 0.5; $CHCl_3$). (ν, cm^{-1}): 1721, 3415, 3573. ^1H spectrum (ppm): 0.84 (s, 3 H), 0.88 (s, 3 H), 0.89 (s, 3 H), 0.96 (s, 3 H), 1.20 (s, 6 H), 1.45 (s, 3 H), 1.52 (s, 3 H), 1.98 (s, 3 H, $OCOCH_3$), 2.09 (s, 3 H, $OCOCH_3$), 3.82 (1 H, t, J = 6.5 Hz, H^{24}), 4.63 (1 H, t, J = 2.7 Hz, H_e^3).

Isolation of the Triterpenes (IX) and (X). Air-dried leaves of *Betula middendorffii* (5 kg), collected at the end of June, 1979, in the environs of the town of Zeya, Amur province, were treated by the method described above. The unsaponifiable fraction of the ethereal extract (26 g) was chromatographed on a column of SiO_2. The triterpene (IX) was eluted with chloroform, and (X) with a mixture of chloroform and ethanol (30:1). Repurification on a column and recrystallization gave the individual triterpenes.

Triterpene (IX). $C_{30}H_{52}O_4$, 500 mg, mp 134-136°C, (acetone).

IR spectrum (ν, cm^{-1}): 1605, 3340, 3420, 3597.

^1H spectrum (ppm): 0.84 (s, 3 H), 0.91 (s, 6 H), 0.94 (s, 3 H), 0.99 (s, 3 H), 1.14 (s, 3 H), 1.28 (s, 3 H), 1.32 (s, 3 H), 3.41 (1 H, t, J = 3.0 Hz, H_e^3), 3.60 (1 H, sextet, $\Sigma J \approx 20$ Hz, H_a^{12}), 5.66 (2 H, broadened singlet, H^{23} and H^{24}).

Mass spectrum (m/e): 458 ($M^+ - H_2O$), 443, 440, 425, 422, 407, 377, 357, 341.

Triterpene (X). $C_{30}H_{52}O_4$, 600 mg, mp 132-135°C (acetone).

IR spectrum (ν, cm^{-1}): 1645, 3300, 3601.

^1H spectrum (ppm): 0.84, (s, 3 H), 0.89 (s, 6 H), 0.94 (s, 3 H), 0.97 (s, 3 H), 1.16 (s, 3 H), 1.72 (s, 3 H, C^{25}—CH_3), 3.40 (1 H, t, J = 2.9 Hz, H_e^3), 3.60 (1 H, m, $\Sigma J \approx 22.0$ Hz H_a^{12}), 4.11 (1 H, t, J = 4.0 Hz, H^{24}), 4.85 (1 H, s, H^{26}), 4.96 (1 H, s, H^{26}).

Mass spectrum (m/e): 458 ($M^+ - H_2O$), 443, 440, 425, 422, 407, 377, 370, 355, 341.

SUMMARY

1. Two new triterpenes have been isolated from the unsaponifiable fraction of an ethereal extract of the roots of *Betula ovalifolia* — 20(S),24(R)-epoxydammarane-3α,17α,35-triol, and 17α,25-dihydroxy-20(S),24(R)-epoxydammaran-3-one.

2. The unsaponifiable fraction of an ethereal extract of the leaves of *Betula middendorffii* has yielded betulafolienetriol and its oxide, and also dammar-23-3n3-3α,12β,20(S),25-tetraol and dammar-25-ene-3α,12β,20(S),24-tetraol.

3. The simultaneous existence of two weakly intramolecular hydrogen bonds has been detected by IR spectroscopy in 17-hydroxy-12-deoxytriterpenoids of the dammarane series with side chains of ocotillol type.

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