COMPOSITION OF THE NEUTRAL FRACTION OF THE RESIN OF PINUS SIBIRICA R. MAYR.

II. Oxygen-Containing Compounds of the High-Boiling Neutral Fraction of the Cedar Resin

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Khimiya Prirodnykh Soedinenii, Vol. 1, No. 4, pp. 250-256, 1965

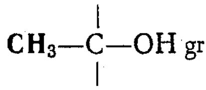
The monoterpene and sesquiterpene compounds of the neutral fraction of the resin of Pinus sibirica R. Mayr. (Siberian cedar) have been studied by Motl, Gerout, and Pentegova [1].

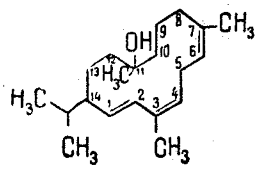
Continuing the investigation of cedar resin, we have found that the high-boiling neutral fraction of the resin contains D-δ-cadinol, which has been obtained previously [1] from the sesquiterpene fraction, and abietinal $C_{20}H_{30}O$. Abietinal was isolated by Erdtman and Westfelt [2] from the extractive substances of Pinus silvestris, and has also been prepared from abietic acid via abietinol [3].

Besides cadinol and abietinal, we have isolated three compounds not described in the literature from the high-boiling neutral substances of the cedar resin: a diterpene alcohol $C_{20}H_{34}O$, a diterpene diol $C_{20}H_{34}O_2$, and a sesquiterpene lactone $C_{17}H_{26}O_3$.

It has been established by hydrogenation that the alcohol $C_{20}H_{34}O$ has three double bonds. The IR spectrum exhibits bands at 850 and 1670 cm^{-1}, which may be assigned to a tri-substituted double bond ($R_1R_2C{=}CHR_3$); at 980 cm^{-1}, to a trans-disubstituted double bond ($-CH{=}CH-$); at 1150 and 3615 cm^{-1}, to a hydroxyl group; and bands at 1370 and 1390 cm^{-1}, to an isopropyl group. The alcohol $C_{20}H_{34}O$ gives no acetic ester on boiling with acetic anhydride, and on treatment with p-nitrobenzoyl chloride or 3, 5-dinitrobenzoyl chloride it dehydrates to the hydrocarbon $C_{20}H_{32}$, which we have identified by a mixed-melting-point test and by comparing the UV, IR, and NMR spectra with cembrene (I).

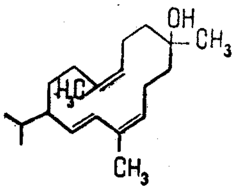
An alcohol isolated with the same carbon skeleton as cembrene, i.e., with a 14-membered ring, is of undoubted interest. Recently seven oxygen-containing compounds with a 14-membered ring contained in tobacco and tobacco smoke have been described [4, 5,6].

The alcohol that we have obtained has been named cembrol. Since under normal conditions cembrol cannot be esterified and is readily converted into cembrene (14-isopropyl-3, 7, 11-trimethylcyclotetradeca-1, 3, 6, 10-tetraene), it may be assumed that it is a tertiary alcohol. The presence in the IR spectrum of the alcohol of maxima at 3615 and 1150 cm^{-1} does not contradict this assumption. The NMR spectrum of cembrol has a proton resonance signal at 8.68τ, which may be assigned to the  $CH_3-\overset{|}{\underset{|}{C}}-OH$ group [5, 6]. The UV spectrum of cembrol shows the conjugation of two double bonds, which is characteristic for cembrene (λ_{max}^{EtOH} 244-246 mμ); consequently, only two positions remain for the tertiary hydroxyl group: on the seventh or eleventh carbon atoms. Hence, structures II and III may be proposed for cembrol.



(II)

II) 14-Isopropyl-3, 7, 11-trimethylcyclotetradeca-1, 3, 6-trien-11-ol

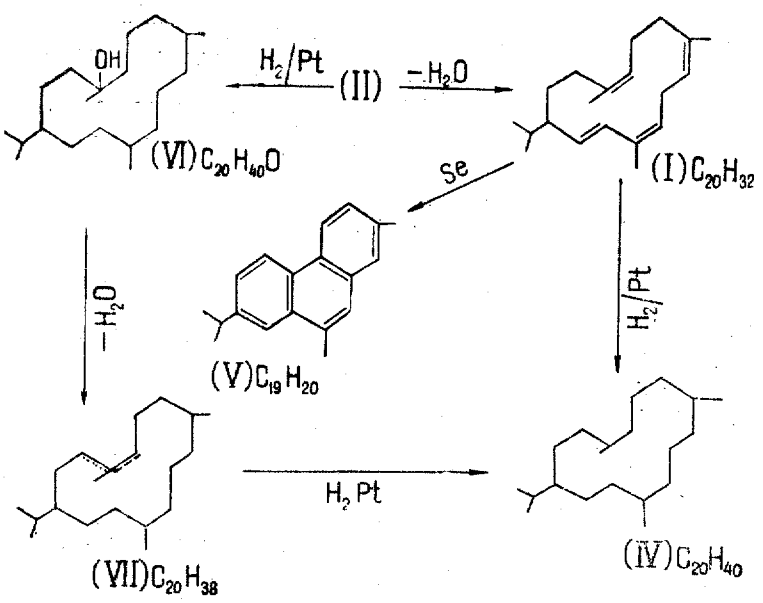


(III)

III) 14-Isopropyl-3, 7, 11-trimethylcyclotetradeca-1, 3, 10-trien-7-ol

Judging from the presence in the NMR spectrum of a proton resonance signal at 7.70τ (which must be assigned to a $=C-CH_2-C=$ group, which does not appear in structure III), II appears the more probable structure.

All the chemical reactions of cembrol mentioned can be represented by the following scheme:



The cembrene (I) obtained by the dehydration of cembrol (II) was dehydrogenated with selenium. The aromatic hydrocarbon isolated was identified as 2, 9-dimethyl-7-isopropylphenanthrene (V) from UV spectra and melting points of the hydrocarbon and its picrate [7, 8] and a mixed-melting-point test.

The hydrogenation of cembrol (II) gave hexahydrocembrol (VI), the dehydration of which gave a hydrocarbon (VII) containing one double bond; when the latter was hydrogenated a saturated hydrocarbon identical from its IR spectrum with octahydrocembrene (IV) was obtained.

The second oxygen-containing compound isolated from the neutral fraction of the cedar resin was a crystalline diterpene diol $C_{20}H_{34}O_2$. From its melting point and specific rotation, this alcohol is similar to the diterpene diol larixol (mp 108-108.5°;$[\alpha]^{20}_{D}$ +33°), which has been isolated from the resin of the European larch in the form of the acetic ester [9, 10] and from the resin of the Siberian larch [11].

However, a mixed-melting-point test of the diol $C_{20}H_{34}O_2$ and larixol (mp 108.5°) gave a depression of the melting point (81-86°). A comparison of the IR spectra of the diol $C_{20}H_{34}O_2$ and larixol also indicated that these two compounds were different.

The third compound obtained from the oxygen-containing neutral fraction of the cedar resin in small amounts consisted of a crystalline substance with mp 79-79.5°. Judging from its elementary composition and its IR spectrum, this compound is apparently a ketolactone $C_{17}H_{26}O_3$.

Experimental

The melting points of the compounds were determined on a Kofler block. The UV spectra were taken in a SF-4 spectrophotometer, and the IR spectra in a UR-10 spectrograph. The mass of the compounds was determined in a MKh-1303 mass spectrograph. The NMR spectra were taken on a JNM-3 instrument (40 mHz). The gas-liquid chromatography was carried out on a "Khrom-1" instrument with a column containing the cyanoethyl ether of sorbitol on INZ-600brick, ratio 20 : 100.

The resin of the Siberian cedar (obtained from the Kebezen Wood-Chemical undertaking, Altai territory) (5340 g) was saponified with 1% caustic soda solution. The neutral unsaponifiables were extracted with diethyl ether. After the ether had been distilled off, 1680 g of neutral substances was obtained, and this was subjected to vacuum distillation with a Vigreux-type fractionating column. 750 g of monoterpene fraction [bp 60-90° (15 mm)] and 450 g of sesquiterpene fraction [bp 90-150° (10 mm)] distilled over. The residue after the distillation (410 g) was chromatographed on alkaline alumina (Brockman activity grade I-II) at a ratio of 1 : 15. This gave 180 g of a diterpene hydrocarbon fraction and 210 g of the oxygen-containing fraction which was the subject of our investigation.

The separation of the mixture of oxygen-containing compounds was carried out on neutral alumina (activity grade II-III) at ratios of 1 : 40 and 1 : 50. Elution was performed with a mixture of petroleum ether and benzene (1 : 1), benzene, a mixture of benzene and diethyl ether (1 : 1), diethyl ether, and a mixture of diethyl ether and ethanol (1 : 1).

The diterpene alcohol – cembrol $C_{20}H_{34}O$. A mixture of petroleum ether and benzene (1 : 1) eluted 51-57% of the total charge of oxygen-containing compounds. Rechromatography on neutral alumina (activity grade II III) at a ratio of 1 : 50 gave 32 g (40-60% of the initial fraction) of cembrol. Its purification was carried out on neutral alumina (activity grade II-III) and on silica gel (KSK, 140 μ, containing 20% of water). The cembrol isolated was a colorless liquid with n_D^{20} 1.5062, $[\alpha]_D^{20}$ +59.6 (c 2.19; chloroform). The content of active hydrogen atoms in it (by Terent'ev's method) was 1.

The IR spectrum of cembrol (in CCl_4) had maxima at 850 cm^{-1}, 900, 920, 940, 980, 1100, 1150, 1370, 1390, 1670, and 3615 cm^{-1}. The NMR spectrum of cembrol on an instrument with a generator frequency of 60 MHz had the following proton resonance signals: doublet at 9.15τ, 868, 850, 840, 8.10, 7.89, 7.70, broad band between 5 and 3.90τ.

Found, %: C 83.12; H 11.84, mol. wt. 290 (mass spectroscopy). Calculated for $C_{20}H_{34}O$, %: C 82.69; H 11.80; mol. wt. 290.

Dehydration of cembrol. A mixture of 1.6 g of cembrol and 1.0 g of p-nitrobenzoyl chloride was heated until the latter had dissolved completely. Cembrene (1.1 g) was obtained from the reaction mixture by recrystallization from ethanol; it had mp (after two recrystallizations) 59-59.5°; $[\alpha]_D^{20}$ +234°(c 1.13; ethanol).

Found, %: C 88.41; H 11.91; mol. wt. 272 (mass spectroscopy). Calculated for $C_{20}H_{32}$, %: C 88.16; H 11.84; mol. wt. 272.

The proton resonance signals in the NMR spectrum of $C_{20}H_{32}$ were: 9.24, 8.60, 8.39, 8.07, and a multiplet centered on 5.08τ. The NMR spectrum of an authentic sample of cembrene taken on the same instrument had signals at 9.26 (isopropyl), 8.62, 8.37 ($CH_3-\overset{|}{C}=$) and 8.06, and a multiplet at 5.08τ, assigned to the olefinic protons.

Hydrogenation of cembrene (I). Cembrene (168.1 mg) was hydrogenated in 10 ml of glacial acetic acid in the presence of 12.6 mg of Adams catalyst. The consumption of hydrogen of 58.1 ml (0°, 760 mm) corresponded to four double bonds (the theoretical consumption of hydrogen for four double bonds is 55.2 ml). The hydrogenation product was extracted in the usual way. This gave 130 mg of octahydrocembrene (IV) with n_D^{20} 1.4798. The test with tetranitromethane was negative.

Found, %: C 86.00; H 14.09. Calculated for $C_{20}H_{40}$, %: C 85.63; H 14.37; mol. wt. 280.

Dehydrogenation of cembrene (I). A mixture of 600 mg of cembrene and 900 mg of selenium was kept at a temperature of 300-330° for 3 hr. The dehydrogenation product, 2, 9-dimethyl-7-isopropylphenanthrene, was purified by chromatography on alkaline alumina (activity grade II-III), mp of the aromatic hydrocarbon 74-76° and of its picrate 133-133.5°; the UV spectrum of the hydrocarbon had maxima at 256 and 298 mμ (log ε 4.84 and 3.898, respectively). An authentic sample of 2, 9-dimethyl-7-isopropylphenanthrene had mp 77.5-78°, mp of the picrate 133°, and maxima in the UV spectrum at 256 and 2.98 mμ (log ε 4.86 and 3.92). A mixture of the two picrates gave no depression of the melting point (133-133.5°).

Hydrogenation of cembrol. Cembrol (2.07 g) was hydrogenated in glacial acetic acid (15 ml) over Adams catalyst (154.8 mg). The consumption of hydrogen was 477 ml (0°, 760 mm); the theoretical amount of hydrogen for three double bonds is 480 ml. The yield of hydrogenation product was 1.64 g (test with tetranitromethane negative). The hydrogenation product was purified by chromatography on silica gel (KSK, 100 μ, air-dried) at a ratio of 1 : 100. A check on the content of substance in the fractions was carried out by means of thin-layer chromatography on a fixed layer of silica gel. A substance with mp 95-97° was obtained. The gas-liquid chromatography of this product gave one peak (cyanoethyl ether of sorbitol, 170°).

The IR spectrum of hexahydroccembrol $C_{20}H_{40}$ (VI) in CCl_4 had bands at 920, 940, 1150, 3615 (hydroxyl), and 1370 and 1390 cm^{-1} (isopropyl group).

Found, %: C 80.98; H 13.35. Calculated for $C_{20}H_{40}O$, %: C 81.00; H 13.60; mol. wt. 296.

Dehydration of hexahydrocembrol (VI). A mixture of 280 mg of hexahydrocembrol and 300 mg of $KHSO_4$ was heated to 170° and was kept at this temperature for 30 min. Then the reaction mixture was extracted with diethyl ether. This gave 220 mg of a brown product which was purified by chromatography on alkaline alumina (activity grade I-II). Petroleum ether eluted 150 mg of a colorless liquid with $n_D^{23.5}$ 1.4840. The test with tetranitromethane gave a yellow coloration. The IR spectrum of the dehydration product (VII) had no bands characteristic for a hydroxyl group, but had bands at 1370 and 1390 cm^{-1} (isopropyl group) and at 920 and 940 cm^{-1}, and a weak maximum at 1675 cm^{-1} which was assigned to the double bond produced.

In the region of weak fields, the NMR spectrum of this hydrocarbon contained a proton resonance signal at 4.98τ which undoubtedly belonged to an olefinic proton.

Found, % : C 86.32; H 13.75. Calculated for $C_{20}H_{38}$, %: C 86.25; H 13.75.

Hydrogenation of the hydrocarbon $C_{20}H_{38}$ (VII). The hydrocarbon (100 mg) was hydrogenated for 2.5 hr under the conditions described previously. The consumption of hydrogen of 10.2 ml (0°, 760 mm) corresponded to one double bond (theoretical amount 9.0 ml). The yield of hydrogenation product was 55 mg, n_D^{20} 1.4796. Test with tetranitromethane was negative.

The IR spectrum of the hydrocarbon was identical with the IR spectrum of octahydrocembrene (IV).

δ-Cadinol. The main component of the fraction eluted with benzene was D-δ-cadinol (80-90% of the fraction), which crystallized out after the solvent had been distilled off. The cadinol isolated (8 g) was recrystallized twice from petroleum ether and was sublimed under vacuum. The melting point of the sublimed substance was 139-140°, $[\alpha]_D^{20}$ +118° (c 3.03; ethanol).

The IR spectrum of the alcohol was identical with that of D-δ-cadinol isolated previously.

Found, %: C 80.79; H 11.71. Calculated for $C_{15}H_{26}O$, %: C 81.02; H 11.79.

The lactone $C_{17}H_{26}O_3$. The mother liquors remaining after the 8 g of cadinol had crystallized out were chromatographed on neutral alumina (activity grade III) at a ratio of 1 : 50, elution with a mixture of benzene and diethyl ether (80 : 20) giving a crystalling substance (80 mg) with mp 79-79.5°.

The IR spectrum of the compound in CCl_4 had maxima at 830 cm^{-1}, 1690 and 3040 cm^{-1} ($R_1R_2C{=}CHR_3$), 890, 1645, and 3085 cm^{-1} ($R_1R_2C{=}CH_2$), 1735 cm^{-1} (C=O), and 1780 cm^{-1} (γ-lactone).

Found, %: C 73.00, 73.10; H 9.20, 8.91; mol. wt. 284 (Rast). Calculated for $C_{17}H_{26}O_3$, %: C 73.34; H 9.41; mol. wt. 278.

Abietinal $C_{20}H_{30}O$. The fraction eluted by a mixture of benzene and diethyl ether (1 : 1) was rechromatographed on neutral alumina (activity grade III, ratio 1 : 50) to give 3 g of a substance with n_D^{20} 1.5264 $[\alpha]_D^{20}$ −72° (c 2.7; ethanol). The UV spectrum of the compound had a maximum at 242 mμ (log ε 3.38). The IR spectrum had bands at 1725 and 2700 cm^{-1} (aldehyde group), and 1670 and 3040 cm^{-1} ($R_1R_2C{=}CHR_3$).

Production of the semicarbazone. A methanolic solution of the aldehyde (470 mg in 2-3 ml of methanol) was mixed with a suspension of semicarbazide hydrochloride (190 mg) and sodium acetate (305 mg) in 5 ml of methanol. The semicarbazone was extracted with diethyl ether. Yield 370 mg. Chromatography of the semicarbazone was carried out on neutral alumina (activity grade III) at a ratio of 1 : 120. Ethanol eluted 302 mg of substance which, after recrystallization from ethanol, had mp 211-212.5°. The IR spectra of the semicarbazones of the aldehyde isolated and abietinal, which we obtained from abietinic acid, were identical.

Found, %: C73.35; H 9.81; N 12.21. Calculated for $C_{21}H_{33}N_3O$, %: C 73.42; H 9.68; N 12.23.

A mixture with an authentic sample of the semicarbazone of abietinal (212-212.5°) gave no depression of the melting point.

The diterpene diol $C_{20}H_{34}O_2$. The main component of the fraction eluted by diethyl ether was a crystalline diterpene diol. After eight recrystallizations from ethyl acetate, 7 g of the crude diol gave 4.6 g of the pure compound (the purity of the substance was checked on plates with a fixed layer of silica gel) with mp 109.5-110.0°, $[\alpha]_D^{20}$ +33° (c 1.83; chloroform). The IR spectrum of the diol in chloroform had maxima at 895, 995, 1035, 1390, 1460, 1480, 1660, 1675, 3040, 3085 and 3640 cm^{-1}; 895, 1660, and 3085 ($R_1R_2C{=}CH_2$). 1035 and 3640 cm^{-1} are assigned to a hydroxyl group.

Found, %: C 78.80; H 11.23; O 10.51. Mass of the compound 288 (306-18) (mass spectrometry). Content of active hydrogen atoms (by Terent'ev's method) 2. Calculated for $C_{20}H_{34}O_2$, %: C 78.40; H 1120; O 10.40; mol. wt. 306. The diol $C_{20}H_{34}O_2$ was insoluble in carbon disulfide, hexane, heptane, and carbon tetrachloride.

Hydrogenation of the diol. The diol (246 mg) was hydrogenated with 30.8 of PtO_2 in absolute ethanol. The amount of hydrogen consumed, 34.5 ml (0°, 760 mm), corresponds to two double bonds (theoretical amount 36 ml). After the ethanol had been distilled off, the hydrogenated product (200 mg) crystallized (mp 98-100°).

The IR spectrum of the saturated diol in CCl_4 had maxima at 1030 cm^{-1}, 1050, 1385, 1460, 3300-3400, and 3635 cm^{-1}.

Found, %: C 77.03; H 12.30. Calculated for $C_{20}H_{38}O_2$, %: C 77.36; H 12.34.

V. A. Koptyug gave advice on the interpretation of the NMR spectra.

Summary

1. The high-boiling neutral fraction of the resin of the Siberian cedar contains abietinal, a diterpene diol $C_{20}H_{34}O_2$, and the diterpene alcohol cembrol $C_{20}H_{34}O_2$ which has a 14-membered ring and three double bonds; the

dehydration of cembrol gives the diterpene hydrocarbon cembrene $C_{20}H_{32}$; the probable structure of cembrol is 14-isopropyl-3, 7, 11-trimethylcyclotetradeca-1, 3, 6-trien-11-ol.

2. Besides the diterpene alcohols and abietinal, the high-boiling oxygen-containing fraction of the cedar resin contains D-δ-cadinol $C_{15}H_{26}O$ and a lactone $C_{17}H_{26}O_3$.

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19 January 1965

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