SUMMARY

The fatty-acid and triacylglycerol compositions of cottonseed forepressing oil and samples of refined oil, salad oil, and palmitin fraction obtained from it have been studied. The type compositions of the diunsaturated-monosaturated triacylglycerols (GlU_2S) of the forepressing, refined, and salad oils were practically identical, while the amount of diunsaturated-monounsaturated (Gl_2SU) the palmitin fraction was far higher than in the others. The salad oil was enriched with triunsaturated triacylglycerols.

LITERATURE CITED

1. P. D. Kupchinsaii, Maslob. Zhir. Promst., No. 2, 8 (1953).
2. A. G. Karantsevich, L. A. Grauerman, and B. Ya. Sterlin, Maslob. Zhir. Promst., No. 6, 41 (1969).
3. D. Tevekelov, Izvestiya na Instituta po Khranene [Bulgaria], 7, 21 (1968).
4. M. G. Kal'nitskii, K. I. Minava, and G. A. Miroslavskaya, Maslob. Zhir. Promst., No. 3, 41 (1969).
5. Handbook on Methods of Investigation, Biological and Chemical Control, and the Accounting of Production in the Oils and Fats Industry [in Russian], Leningrad, Vol. 1, Book 2 (1967).
6. T. V. Chernenko, A. L. Markman, and A. U. Umarov, Prikl. Biokhim. Mikrobiol., No. 3, 616 (1969).

TERPENOIDS FROM PLANTS OF THE FAMILY CUPRESSACEAE.

I. SESQUITERPENE ALCOHOLS FROM THE NEEDLES OF *Microbiota decussata*

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The sesquiterpene alcohols 5S,8S-germacra-1E,6E-dien-5-ol, (+)-α-bisabolol, hedycaryol, and β-eudesmol and also the previously undescribed alcohols thujopsan-2α-ol (I) and microbiotol. Microbiotol is a tricyclic tertiary alcohol with the empirical formula $C_{15}H_{26}O$, mp 112-113°C, containing in its molecule four tertiary methyl groups and cyclopropane ring.

Microbiota (*Microbiota decussata* Kom.) belongs to the only endemic monotypical genus of conifers of the USSR and is distributed in the Sikhote-Alin' range in Maritime Territory and in the south of the Khabarovo Territory [1]. This monoecious evergreen bush of the family Cupressaceae has its maximum distribution in the southern part of the Sikhote-Alin' range (Partizanskii and Livadiiskii ranges) and here it grows en masse, covering the rocky alluvial deposits completely or partially. Usually, microbiota forms a band above the forest vegetation, but not infrequently along the mountain springs through the breaks it comes down from the high-mountain region to a height of 300 m above sea level, and at the edges of the rocky deposits it is sometimes found below the forest threshold. In the northern part, the area of microbiota is disjunctive. Separate growth sites of the species have been reported in the upper course of the river Khor and on the watershed between the rivers Khor and Anyui in the Khabarovo Territory. In Komarov's opinion [2], the tree and the generalized genus *Microbiota* Kom. are closest to the genus *Biota*.

This plant has not previously been studied chemically. For the investigation we used fresh needles collected in the autumn. An etheral extract obtained by steeping at room tem-

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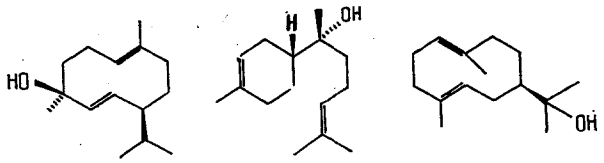
temperature (yield about 6% on the absolute dry weight of the needles) was chromatographed on silica gel. A mixture of hydrocarbons, wax-like substances, and fractions containing individual substances, called in order of the elution substances A, B, C, D, and E, were eluted successively. According to their IR and NMR spectra, all the substances were tertiary alcohols.

On the basis of its constants, spectral characteristics, and a comparison by TLC with an authentic sample, alcohol A was identified as 5S,8S-germacra-1E,6E-dien-5-ol (I), which has been isolated previously from the oleoresin of the Yeddo spruce [3]. Alcohol B was identified similarly as (+)-α-bisabolol (II). This compound is fairly widely distributed in plants [4], both its enantiomers having been found in nature. Schwartz and Swanson [5] have recently effected a stereospecific synthesis of α-bisabolol which has permitted the stereochemistry of this compound to be refined.

Alcohol C is adsorbed strongly on silica gel impregnated with silver nitrate, which shows the strength of its complex with silver ions. This has been used for the direct isolation of alcohol from the initial mixture of alcohols under investigation. When a hexane solution of this mixture was treated with 20% aqueous silver nitrate solution, alcohol C passed completely and selectively into the aqueous solution, the dilution of which with hot water and extraction with diethyl ether yielded a pure sample of alcohol C in the form of an oil. On the basis of the constants and the results of IR and NMR spectroscopy, it was identified as hedycaryol (III) which has been isolated previously from three species of plants [6, 7].

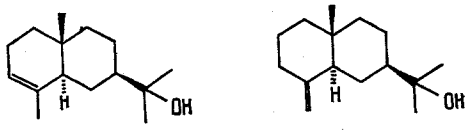


I II III

Hedycaryol is a precursor of the well-known sesquiterpene alcohol elemol, into which it is converted at a high temperature through the occurrence of a Cope rearrangement [6]. We detected no elemol in the extract investigated, but when the needles were extracted in Soxhlet apparatus followed by treatment of the extract with silver nitrate, we obtained a sample of hedycaryol containing, according to its NMR spectrum, about 20% of elemol.

As Jones and Sutherland [6] have established, when an ethereal solution of hedycaryol is boiled with p-toluene sulfonic acid, a mixture of α-, β-, and γ-eudesmols is formed. We found that hedycaryol isomerizes into a mixture of α- and β-eudesmols (IV and V, respectively; ratio 2:3) even when its solution in carbon tetrachloride is allowed to stand at room temperature. The course of isomerization is easily followed from the change in the integral intensities of the signals of the olefinic protons in the NMR spectrum. The transformation takes place almost completely in three days.

In the initial needle extract, β-eudesmol was found in very small amount, and it was isolated in the process of prolonged chromatographic purification of substance D on silica gel containing silver nitrate. The amount of β-eudesmol in the extract rose when the extract was stored at room temperature for a long time or when it was evaporated at an elevated temperature (60-70°C).



IV V

Eudesmols have been isolated from a number of plants [7, 8]. However, the ease of conversion of hedycaryol into eudesmols permits the assumption that these compounds may be secondary products formed from hedycaryol through severe conditions of isolation or treatment of the raw material (steam distillation and melting point).

Substance D proved to be a previously undescribed compound. It was obtained in the form of colorless prisms with mp 57-58°C (from ethylene). The hydroxy group the presence of

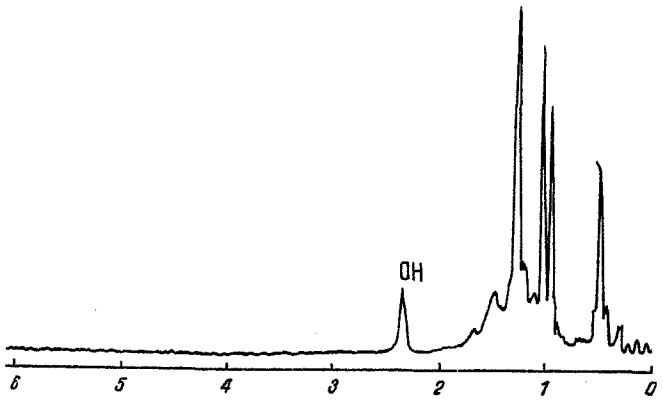


Fig. 1. NMR spectrum of thujopsan-2α-ol (VII) (60 MHz).

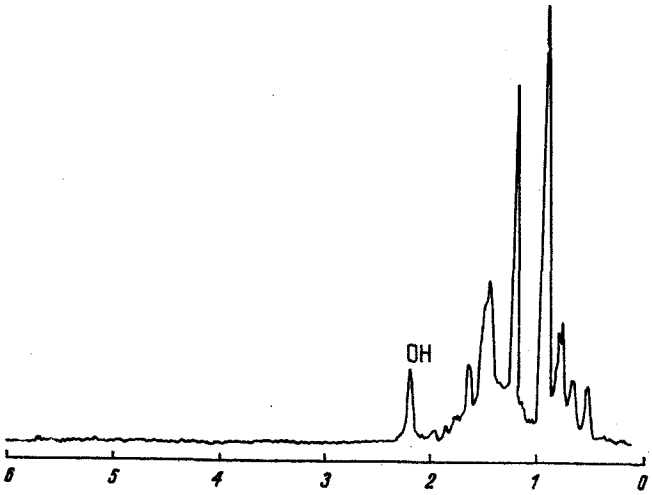
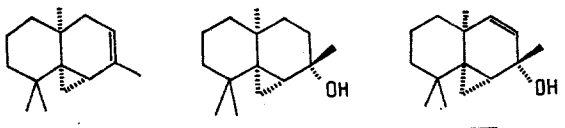


Fig. 2. NMR spectrum of microbiotal (60 MHz).

which in substance D was established from its UV spectrum (3620 cm^{-1}) is tertiary, since the NMR spectrum (Fig. 1) lacks the signals of carbinol protons and has the signal of a tertiary methyl group (1.28 ppm) geminal to a hydroxy group. Furthermore, in the NMR spectrum the singlet signals of three other tertiary methyl groups are observed (1.03, 0.96, and 0.50 ppm), one of which (0.50 ppm) is strongly screened by a cyclopropane ring the presence of which is confirmed by the IR spectrum (3060, 3040, 920 cm^{-1}) and by signals in the 0.3-0.5 ppm region of the NMR spectrum. In the high-resolution mass spectrum the molecular ion is not observed, and the peak of the ion with highest mass corresponds to the formula $C_{15}H_{24}$, which permits its interpretation as $(M - H_2O)^+$.

When substance D was degraded with phosphorus oxychloride in pyridine at 0°C, a hydrocarbon was formed which was identified by its constants and spectral characteristics as (−)-thujopsene (VI). The ^{13}C NMR spectrum of substance D lacked the signals of carbon atoms bound by double bonds. In view of the results obtained, it may be concluded that substance D is thujopsan-2-ol (VII). The intermediate formation of this compound was suggested by Dauben and Ashcraft [9] in the addition of methylmagnesium iodide to northujopsan-2-one.



VI VII VIII

The configuration of the asymmetric center of thujopsan-2-ol at C_2 was determined in the following way. Δ^3-Thujopsen-2α-ol (VIII), obtained by the photooxidation of (−)-thu-

jopsene by Ito's method [10], on hydrogenation in ethanol over platnum dioxide gave thujopsan-2α-ol (VII), identical to the alcohol D.

The high stereospecificity of the dehydration of thujopsan-2-ol to thujopsene permits the assumption that the hydroxy group in thujopsan-2-ol is present in the pseudoaxial configuration. This can apparently explain the instability of thujopsan-2-ol. Even at room temperature in chloroform or carbon tetrachloride solutions it gradually dehydrates to (−)-thujopsene. It is possible that the (−)-thujopsene isolated from many plants of the family Cupressaceae [11] is formed, even if partially, by such a nonenzymatic dehydration of thujopsan-2-ol.

Alcohol E, called microbiotol, also proved to be a new, previously undescribed, compound. It was obtained in the form of colorless fine acicular crystals with a specific sweetish smell, readily subliming in vacuum. Its volatility was used to separate the sterols that were eluted from the column together with it in chromatograph. The elementary analysis of microbiotol corresponded to the empirical formulas $C_{15}H_{26}O$, but in the high-resolution mass spectrum only the peaks of ions corresponding in composition to the product of the splitting out of a molecule of water from the molecular ion ($C_{15}H_{24}^{+}$) were observed. The UV spectrum of microbiotol has no absorption maxima in the 220-400 nm region, and in the IR spectrum the absorption bands of a hydroxy group (3620 cm^{-1}) and of a cyclopropane ring (3075, 975, 945, 920 cm^{-1}) were observed. The NMR spectrum (Fig. 2) shows the signals of four tertiary methyl groups, three of which have the same chemical shift (0.95 ppm, 9 H), and one, being geminal to the hydroxy group, giving a singlet at 1.25 ppm. In addition, in the 0.52-0.7 ppm region signals are observed that can be assigned to the protons of a cyclopropane ring. Since in the ^{13}C NMR spectrum of microbiotol there are no signals corresponding to carbon atoms at double bonds, then, on the basis of the results obtained, it may be concluded that microbiotol is a tricyclic saturated tertiary alcohol one of the rings of which is a cyclopropane ring. The study of the structure of this compound is continuing.

In the combined sesquiterpene alcohols of micriobiota needles, the hedycaryol amounted to about 47%, thujopsenol to 35%, microbiotol to 9%, germacra-1E,6E-dien-5S-ol to 6%, the bisabolol to 3%, and β-eudesmol to about 0.5%.

In the sample of microbiotol needles investigated, we detected no appreciable amount of diterpene resin acids and neutral diterpenoids, which are characteristic for plants of the Pinaceae [12].

EXPERIMENTAL

For the investigation we used fresh needles of microbiota collected in the autumn of 1978 in the Shkotovo region of the Maritime Territory.

Melting points were determined on a Kofler block, and angles of rotation were measured for solutions in chloroform on a Ziess polarimeter. PMR spectra (δ scale, internal standard hexamethyldisiloxane) were recorded on Varian A56/60A and Varian HA-100 instruments. IR spectra were taken for solutions in carbon tetrachloride on a UR-20 instrument, and mass spectra on an MS-902 spectrometer. ^{13}C NMR were obtained on a Bruker HX-90 pulsed spectrometer (22.63 MHz) for solutions in deuterochloroform.

For chromatography we used air-dry KSK silica gel with a grain size of 0.140-0.315 mm, and the same silica gel containing 5% of silver nitrate. As the eluent in all cases we used petroleum ether with increasing amounts (from 0 to 40%) of diethyl ether.

Extraction of the Initial Raw Material. Ground fresh microbiota needles (200 g) were covered with one liter of diethyl ether and left at room temperature for 24 h, after which the extract was decanted off and a new portion (1 liter) of diethyl ether was poured on. The combined extracts were evaporated to 0.5 liter and were washed with 200 ml of saturated aqueous sodium bicarbonate, and the solvent was distilled off. This gave 5 g of extract in the form of a waxy mass readily soluble in diethyl ether and only partially soluble in hexane.

Isolation of the Total Sesquiterpene Alcohols. A solution of 5.0 g of the needle extract in 15 ml of diethyl ether was mixed in a porcelain dish with 20 g of silica gel and was left in the air until the diethyl ether had evaporated off completely. The resulting powder was transfered to a column filled with 200 g of silica gel. Petroleum ether eluted

1.2 g of hydrocarbons from the column, petroleum ether containing 5% of diethyl ether eluted 1.0 g of a mixture of waxy substances, and petroleum ether containing 20% of diethyl ether eluted 2.1 g of a mixture of terpenoid alcohols. On further elution of the column, chlorophyll and more polar compounds were obtained.

Hedycaryol (III). A solution of the 2.1 g of terpenoid alcohols in 20 ml of hexane was treated with 20% aqueous silver nitrate as described by Jones and Sutherland [6]. This yielded 1.2 g of a mixture of alcohols not forming a complex with silver nitrate and 0.8 g of hedycaryol with n_D^{20} 1.5128 and $[\alpha]_D^{21}$ +30.5° (c 4.5), p-nitrobenzoate with mp 111-112°C (from carbon tetrachloride); according to the literature [6]: $[\alpha]_D^{25}$ +30.8°, p-nitrobenzoate with mp 110-112°C.

Isomerization of Hedycaryol. A solution of 0.1 g of hedycaryol in 0.5 ml of deuterochloroform containing about 0.01 g of hexamethyldisiloxane was left at room temperature for 3 days. A gradual disappearance of the signals of the olefinic protons of the hedycaryol in the NMR spectrum (multiplet at 4.83 ppm) and the appearance of signals corresponding to α- and β-eudesmols (5.20 and 4.38, 4.60 ppm, respectively) was observed. Chromatography of the product on silica gel containing 5% of silver nitrate yielded 0.03 g of α-eudesmol with mp 81-82°C and $[\alpha]_D^{20}$ +20.5° (c 2.0) (according to the literature [8b] mp 81-82°C $[\alpha]_D^{25}$ +30.5°) and 0.05 g of β-eudesmol with mp 79-80°C (sublimed in vacuum) and $[\alpha]_D^{20}$ +58.8° (c 3.0) (according to the literature [13]: mp 79-80°C). The NMR spectra of the eudesmols corresponded to those published [8a].

Separation of the Combined Sesquiterpene Alcohols not forming Complexes with Silver Nitrate. The mixture of alcohols (1.2 g) remaining after the separation of the hedycaryol was chromatographed on 40 g of silica gel. This led to the successive elution of 0.1 g of 5S,8S-germacra-1E,6E-dien-5-ol (I) with $[\alpha]_D^{20}$ −176° (c 9.09) and n_D^{20} 1.4958 (according to the literautre [3]: $[\alpha]_D^{22}$ −177°, n_D^{22} 1.4961), 0.05 g of (+)-α-bisabolol (II) with $[\alpha]_D^{22}$ +49.4° (c 3.85) and n_D^{22} 1.4915 (according to the literature [14]: $[\alpha]_D$ +43° and n_D^{24} 1.4908), 0.61 g of crude thujopsanol and 0.3 g of crude microbiotol.

The rechromatography of 0.61 g of the crude thujapsanol on 20 g of silica gel containing 5% of silver nitrate yielded 0.60 g of pure thujopsan-2α-ol (VII) with mp 57-58°C (from hexane) and $[\alpha]_D^{21}$ −29.6° (c 10.8), and 0.05 g of β-eudesmol with mp 79-80°C, giving no depression of the melting point with the sample of β-eudesmol obtained by the isomerization of hedycaryol.

The crude microbiotol was freed from impurities by sublimation in vacuum (1 mm Hg) at 70-80°C. This yielded 0.15 g of microbiotol in the form of very thin acicular crystals which, after recrystallization from acetonitrile, had mp 116-117°C and $[\alpha]_D^{25}$ −13° (c 3.08). The residue from sublimation (about 0.13 g) was practically completely acetylated by acetic anhydride in pyridine at room temperature, forming a product coinciding on TLC with a sample β-sitosterol acetate.

(−)-Thujopsene (VI). At 0°C, 1 ml of phosphorus oxychloride was added dropwise to a solution of 0.5 g of thujopsan-2α-ol in 10 ml of pyridine, and was left at 0°C for 24 h. After the usual treatment and chromatography on silica gel (eluent – petroleum ether), 0.3 g of (−)-thujopsene (VI) was obtained with n_D^{20} 1.5040 and $[\alpha]_D^{21}$ −108° (c 7.88) according to the literature [11], n_D^{25} 1.5031, $[\alpha]_D$ −110°. The IR spectrum of the sample of thujopsene obtained coincided completely with that given by Wenninger et al. [15].

Synthesis of Thujopsan-2α-ol. Δ^3-Thujopsen-2α-ol (VIII) (0.2 g), obtained by the photooxidation of (−)-thujopsene by Ito's method [10] was hydrogenated in 20 ml of ethyl acetate over 0.1 g of platinum dioxide. After chromatography of the product on 15 g of silica gel, 0.15 g of thujopsan-2α-ol (VII) was obtained with mp 56-58°C; its IR and NMR spectra agreed with those for the sample of thujopsan-2α-ol isolated from the needles under investigation.

SUMMARY

1. The needles of microbiota (*Microbiota decussata*. Kom.) have yielded six sesquiterpene alcohols – 5S,8S-germacra-1E,6E-dien-5-ol (+)-α-bisabolol, hedycaryol, β-eudesmol, thujopsan-2α-ol, and microbiotol.

2. The structure and stereochemistry of a new sesquiterpene alcohol – thujopsan-2α-ol – has been established on the basis of spectral and chemical characteristics.

LITERATURE CITED

1. N. N. Gurzenkov and P. G. Gorovoi, collection: Endemic High-Mountain Plants of Central Asia [in Russian], Novosibirsk (1974), p. 5; P. G. Gorovoi and D. D. Basargin, in: A List of Plants in the Herbarium of the Flora of the USSR [in Russian], Leningrad, No. 22 (1979), p. 46.
2. V. L. Komarov, Botanical Materials of the Herbarium of the Main Botanical Garden [in Russian], Vol. 4 (1923), p. 23.
3. V. A. Raldugin, V. L. Salenko, N. S. Gamov, T. F. Titova, V. A. Khan, and V. A. Pentegova, Khim. Prir. Soedin., 199 (1980).
4. F. Sorm, M. Zaoral, and V. Herout, Collect. Czech. Chem. Commun., 16, 626 (1951); F. Sorm, M. Vrany, and V. Herout, Chem. Listy, 46, 364 (1952).
5. M. A. Schwartz and M. D. Swanson, J. Org. Chem., 44, 953 (1979).
6. R. V. Jones and M. D. Sutherland, J. Chem. Soc., Chem. Commun., 1229 (1968).
7. a) I. A. Southwell, Phytochemistry, 9, 2243 (1970). b) I. A. Southwell, Aust. J. Chem., 31, 2527 (1978).
8. a) H. P. Korthals, D. Merkel, and M. Muhlstadt, Ann. Chem., 745, 39 (1971). b) F. J. McQuillin and J. D. Parrack, J. Chem. Soc., 2973 (1956).
9. W. G. Dauben and A. C. Ashcraft, J. Am. Chem. Soc., 85, 3673 (1963).
10. S. Ito, H. Takeshita, T. Muroi, M. Ito, and K. Abe, Tetrahedron Lett., 3091 (1969).
11. T. Norin, Acta Chem. Scand., 15, 1676 (1961).
12. V. A. Raldugin, V. A. Khan, Zh. V. Dubovenko, and V. A. Pentegova, Khim. Prir. Soedin., 609 (1976).
13. H. Yoshioka, T. J. Mabry, and A. Higo, J. Org. Chem., 34, 3697 (1969).
14. V. A. Babkin, Zh. V. Dubovenko, and V. A. Pentagova, Izv. Sibirsk. Otd. Akad. Nauk SSSR, Ser. Khim. Nauk, series No. 1, Issue No. 2, 168 (1970).
15. J. A. Wenninger, R. L. Yates, and M. Dolinsky, J. Assoc. Offic. Anal. Chem., 50, 1313 (1967).

AN EMPIRICAL CORRELATION BETWEEN STRUCTURE AND OPTICAL ACTIVITY IN A SERIES OF LABDANE DITERPENOIDS

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On the basis of the results of an analysis of the optical rotatory dispersion curves and a consideration of possible conformations of labdane diterpenoids, the conclusion has been drawn that labd-8(17)-ene derivatives with a 13E double bond or a voluminous substituent in the side chain do not obey Garman's additive scheme, and when there are no asymmetric centers in the side chain they have more negative values of the molecular optical rotation than is predicted by this scheme.

On the basis of an analysis of the molecular optical rotations of a large number of labdane diterpenoids, Carman [1] put forward an additive scheme for calculating these magnitudes for new derivatives of the labdane series. According to the scheme, the optical rotation at 589 nm (sodium D line) of a labdane derivative is considered as the sum of the contributions of two moieties of the molecule of a compound with a labdane carbon skeleton (I) — the aliphatic (C_{11}—C_{16}) and the cyclic (C_1—C_{10}). This scheme is based on the assumption of the free rotation of the aliphatic fragment of the molecule around the C_9—C_{11} and C_{11}—C_{12} bonds. When there are no asymmetric centers in the aliphatic part of the molecule, it is considered that the observed optical rotation is due only to the cyclic fragment, which contains at least three asymmetric centers (at C_5, C_9, and C_{10}).

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