LABDANE ACIDS AND OTHER COMPONENTS OF THE NEEDLES OF *Pinus pumila*

V. A. Raldugin, L. I. Demenkova,
and V. A. Pentegova

UDC 547.595.9

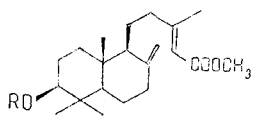
The terpenoid and aliphatic components of an ethereal extract of the oleoresin of the Japanese stone pine have been investigated. Among them, 33 compounds have been identified, ten of which were isolated in the individual state. The labdane acids of the oleoresin investigated were represented by 19-O-succinylagatholic and 3β-hydroxy-, 3β-acetoxy, and 3-ketoanticopalic acids.

The Japanese stone pine covers a fairly wide area in the eastern regions of the USSR [1], in view of which it may be considered as potential chemical raw material. Its oleoresin has been little studied and therefore great interest from the practical point of view is presented by the needles of the species, about which there is only a mention by Zinkel [2] in the literature of the high content of neoabietic acid in them. Although this species of *Pinus* L. is included, according to Mirov's classification [3], in the same botanical group (*Cembrae*) as the Siberian, Korean, and Swiss stone pines (*P. sibirica* R. Mayr., *P. koraiensis* Sieb. et Zucc., and *P. cembra* L.) it is sharply distinguished from the latter by the composition of the diterpenoids of the oleoresin [4-6].

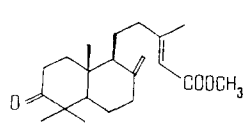
We have investigated the components of an ethereal extract of the air-dry resin collected in August, 1983, in the Kurile islands. It was possible to perform a comparatively complete analysis of the terpenoid components of the extract obtained by using a scheme for the group separation of the components of conifer oleoresins [7]. In order to simplify the analysis of the acid components of the extract, its ethereal solution was treated with saturated aqueous solution of sodium bicarbonate. This operation permitted the selective isolation from the total of the so-called "strong" acids [6]. The subsequent treatment of the ethereal solution with a 2% aqueous solution of caustic soda isolated all the remaining acids, and, after the solvents had been eliminated, a neutral fraction was obtained. By this scheme, from 633 g of oleoresin 26.40 g (4%) of extract was obtained from which 2.73 g of strong acids, 10.61 g of other acids, and 13.00 g of a neutral fraction (10.3, 40.1, 49.2%, respectively, on the extract) were isolated.

The strong-acid fraction consisted of two components with traces of other compounds. The main components — benzoic and 19-O-succinylagatholic acids — were isolated by chromatography in the form of their methyl esters and were identified from their IR and PMR spectra. Benzoic acid — the main component of this fraction (~85%) has not previously been detected in appreciable amounts in the needles of the Siberian stone pine [8, 9]. 19-O-Succinylagatholic acid is one of the components of the strong-acid fraction of the oleoresin of the Japanese stone pine [6].

The acidic part of the extract isolated by treating the solution with caustic soda was methylated with an ethereal solution of diazomethane and the products were chromatographed on silica gel. This gave a mixture of methyl esters of resin acids (in a yield of 48% on the total amount of methyl esters taken) and the methyl esters of 3β-acetoxyanticopalic acid (I) (24.6%), of 3-ketoanticopalic acid (II) (8.9%), and of 3β-hydroxyanticopalic acid (III) (9.2%).



I R = $COOCH_3$
III R = H



II

Novosibirsk Institute of Organic Chemistry, Siberian Branch, Academy of Sciences of the USSR. Translated from Khimiya Prirodnykh Soedinenii, No. 2, pp. 206-211, March-April, 1985. Original article submitted May 3, 1984.

 0009-3130/85/2102-0192$09.50

3β-Hydroxy- and 3β-acetoxyanticopalic acids have been found previously in the oleoresin of the Japanese stone pine [6]. 3-Ketoanticopalic acid was isolated by Zinkel [10] from the needles of *Pinus strobus* L., but he did not give the spectral characteristics and physical constants of this acid or its methyl ester, nor a proof of its structure. In this connection, we have synthesized the keto ester (II) by oxidizing the hydroxy ester (III) with the Jones reagent in acetone. The product obtained proved to be identical with the natural product according to its spectral, TLC, and optical properties.

We did not succeed in detecting agatholic (19-hydroxyanticopalic) acid and its 19-O-acetate, which are present in the oleoresin of the Japanese stone pine [6]. Nevertheless, it must be mentioned that the biological oxidation of anticopalic acid at C-19 in the needles of the Japanese stone pine does actually take place, as is shown by the presence of 19-O-succinylagatholic acid in it. The main direction of the metabolism of anticopalic acid here is oxidation at C-3.

At room temperature, the mixture of methyl esters of resin acids consisted of a semisolid waxy mass. Such an external form permitted the assumption that the methyl esters of higher fatty acids were present in it. In actual fact, the sum of these esters was obtained with a yield of 10% on the fraction taken by chromatography on silica gel impregnated with silver nitrate. According to GLC, it was a mixture of the methyl esters of the normal fatty acids with even numbers of carbon atoms in the molecule — from C_{12} (lauric) to C_{24} (lignoceric) (the amounts in the mixture being 4, 7, 26, 5, 12, 15, and 31%, respectively) with traces of other, unidentified, esters.

According to GLC, the sum of the resin acid methyl esters consisted of the esters of sandaracopimaric (13.8%), isopimaric (8.3%), palustric + levopimaric (23.6%), dehydroabietic (11.8%), abietic (4.8%) and neoabietic (37.7%) acids. According to the PMR spectra of the sum of the methyl esters obtained, it included in equal amounts both methyl palustrate and methyl levopimarate. The high proportion of sandaracopimaric acid is unusual for species of *Pinus* L. Its methyl ester, isolated by chromatographing the combined esters on silica gel impregnated with silver nitrate had a PMR spectrum coinciding with that given in the literature [11]. Anticopalic acid — the main acid of the oleoresin of the Japanese stone pine [4] was not detected in the needles investigated.

When the neutral fraction of the extract was diluted with 3 volumes of hexane, and cooled, it deposited a waxy substance (21.9%; here and below the yields are given in relation to the initial neutral fraction of the extract). This product, according to its PMR and IR spectra, was a mixture of saturated esters of fatty acids and alcohols with free fatty alcohols. By chromatographing the mother liquor on silica gel in accordance with the scheme of group separation [7], the total hydrocarbons (42.5%), a fraction of carbonyl compounds (8.0%), a fraction including all the substances then eluted up to the polar monohydric alcohols (8.0%), a fraction of polar monohydric alcohols (8.6%), and the combined polar compounds (10.5%) were obtained.

The hydrocarbons, according to GLC, consisted of monoterpenes (66.4%), sesquiterpenes (29.8%), diterpenes, and higher paraffins (3.8%). Their quantitative composition was not studied.

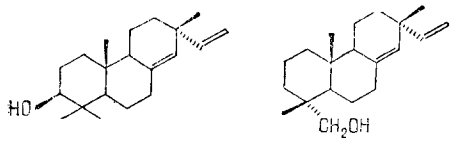
Treatment of the fraction of carbonyl compounds with sodium tetrahydroborate [7] gave alcohols from the aldehydes (2%) and a mixture of esters (5%). The alcohols from the aldehydes were converted into the acetates by treatment with acetic anhydride in pyridine and were analyzed by the GLC method with the addition of authentic samples. It was established that the sum of the aldehydes of the needles investigated consisted of sandaracopimarinal (26.2%), palustral (21.2%), dehydroabietinal (11.3%), abietinal (20.9%), and neoabietinal (20.5%). The correctness of the identification of the alcohols from the aldehydes, which was made by GLC, was confirmed by the PMR spectrum (90 MHz) of a mixture of their acetates, in which the signals of olefinic protons of all the compounds identified were observed in the 4.5-7.2 ppm region.

The fraction of esters obtained after the elimination of aldehyde from it consisted of the methyl esters of resin acids (one quarter) and of a mixture (three quarters) of esters of higher fatty acids and alcohols (estimated from the ratio of the signals of the protons from the $COOCH_3$ and $-CH_2-OCO$ groups which were observed in the PMR spectrum at 3.64 ppm (singlet) and 4.04 ppm (triplet, J = 6 Hz), respectively). According to GLC, the combined methyl esters contained as the main components methyl abietate (34.0%), methyl dehydroabietate (25.2%), methyl neoabietate (15.8%), and methyl sandaracopimarate (6.8%).

The fraction eluted from the column after the carbonyl compounds was a complex mixture of substances in which no main components whatever were expressed. When its solution in hexane was kept in the refrigerator, crystals of nonacosan-10-ol, a common component of conifer needles [8, 12], deposited.

Treatment of the fraction of polar monohydric alcohols by scheme [7] gave the combined acetates of the acetylatable alcohols (6.2%) and the tertiary alcohols (1.5%). The latter were represented by α-cadinol, with traces of other compounds. The α-cadinol was identified by TLC and by IR and PMR spectroscopy. Chromatography of the acetates fraction on silica gel impregnated with silver nitrate permitted the isolation of its main component (~90% of the fraction), which was reconverted into the alcohol by reduction with lithium tetrahydroaluminate in diethyl ether. The alcohol obtained had mp 126-127°C and $[\alpha]_D^{20}$ −12.66° (c 13.42). Its PMR spectrum (60 MHz) was similar to that given in the literature for enantiosandaracopimaradien-3β-ol (enantioisopimara-8(14),15-dien-3β-ol) (mp 126-127°C, $[\alpha]_D$ + 12.5°), isolated by Candy et al. [13] from the heartwood of *Cleistanthus schlechteri*.

Sandaracopimaradien-3β-ol (IV) itself, obtained by Laidlaw and Morgan [14] from the wood of *Xylia dolabriformis*, has mp 126.5-127.5°C, $[\alpha]_D$ − 19.5°. By using sandaracopimarinol (V) as marker, it was established that the latter in the free form and its acetate were chromatographically indistinguishable from the alcohol (IV) and its acetate, respectively. In this connection, the question arises of the homogeneity of samples of the alcohol (IV) obtained on chromatographic separation. It was found that it was easily answered by recording the PMR spectrum on an instrument with a working frequency of 200.13 MHz. When this spectrum was recorded for the sample of the alcohol (IV) that had been isolated it was established that it was a mixture of the isomers (IV) and (V) in a ratio of 5:1 (from the relative integral intensities of the separately observed signals of carbinol protons — H_3 for (IV) (doublet of doublets at 3.21 ppm, J = 11.0 and 4.5 Hz), and $-\underline{CH_2}OH$ for (V) (AB system with components at 3.05, 3.10, 3.33, and 3.38 ppm, J_{AB} = 11.0 Hz)).



IV V

The signals of the olefinic protons and of the methyl groups of sandaracopimarinol (V) were superposed on the corresponding signals of the alcohol (IV). In an instrument with a working frequency of 60 MHz, the signals of the carbinol protons in the spectrum of the mixture of these compounds formed a single broad multiplet, as described in the literature [13] for the alcohol (IV).

The incomplete oxidation of a sample of sandaracopimaradien-3β-ol, the constants of which corresponded to those given in the literature [13], yielded a mixture of sandaracopimarinal, 3-ketosandaracopimaradiene, and sandaracopimaradien-3β-ol, which was readily separated by chromatography on silica gel. The pure sandaracopimaradien-3β-ol (IV) obtained from this mixture had mp 129-130°C and $[\alpha]_D^{17}$ − 22.7 (c 3.52). The sandaracopimarinal was identified by its PMR spectrum and by conversion into sandaracopimarinol, identical with an authentic sample according to its PMR spectrum [15].

Thus, the needles investigated contained the two alcohols (IV) and (V) in a ratio of 5:1. The acetylatable alcohols accompanying them were represented by equal amounts of neoabietinol, abietinol, dehydroabietinol, palustrol and (or) levopimarinol (GLC).

The fraction of polar compounds (10.5% of the initial neutral part of the extract of the needles) contained one main component, which was purified in the form of its acetate (yield ~ 8%). It was identified from its constants and spectral characteristics as sandaracopimaradiene-3β,18-diol, which has been isolated previously from the wood of *Xylia dolabriformis* [14].

The high content of sandaracopimaradiene derivatives in the stone pine needles, and of anticopalic acid derivatives, is a feature of this plant. Compounds of such a type are quite uncharacteristic for plants of the family Pinaceae, and the appreciable predominance of this direction of biosynthesis of the isopimarane diterpenoids additionally [6] shows the noncorre-

spondence of the positions of the Japanese stone pine in the same botanical group (*Cembrae*) as other species of *Pinus* L.

EXPERIMENTAL

PMR spectra were recorded on Varian A 56/60A (60 MHz), Bruker HX-90 (90 MHz) and Bruker WP-200 SY (200.13 MHz) instruments using solutions in carbon tetrachloride (with MHDS as internal standard) and in deuterochloroform (using chloroform as internal standard, the signal from which was taken as 7.24 ppm), δ scale. IR spectra were obtained on a UR-20 instrument. Angles of optical rotation were measured for solutions in chloroform on a Zeiss polarimeter. Gas-liquid chromatography was performed on a Chrom-41 instrument (Czechoslovakia) with a 2.5 m × 3 mm glass column containing as stationary phase 5% of SE-30 on Chromaton N-AW, DMCS (0.200-0.250 mm). The temperature of the column was 90-240°C/2°C for the hydrocarbons, and 210°C for the alcohol acetates. For the analysis of the methyl esters of the resin acids we used the same column but as stationary phase OV-225 on Chromaton N-Super (0.160-0.200 mm) at a column temperature of 210°C. The methyl esters of the fatty acids were analyzed with the use of the stationary phase 5% PDEGS + 1% OV-225 on Chromaton-N-AW, DMCS (0.200-0.250 mm) at a column temperature of 130-220°C/3°C.

For adsorption chromatography we used air-dry type KSK silica gel with a grain diameter of 0.140-0.315 mm, the eluent in all cases being petroleum ether (bp 40-70°C) with increasing (from 0 to 100%) concentrations of diethyl ether, the ratio of substance and sorbent being ~1:30.

Isolation of the Main Groups of Compounds from Japanese Stone Pine Needles. The air-dry Japanese stone pine needles (633 g) were extracted with diethyl ether (2 liters) in a Soxhlet apparatus for 4 h. Evaporation of the extract gave 26.40 g of product, which was dissolved in 0.5 liter of diethyl ether and treated with a saturated aqueous solution of sodium bicarbonate (300 ml). Acidification of the aqueous bicarbonate solution to pH 2 with hydrochloric acid and extraction with diethyl ether gave the total strong acids (2.73 g). The residual ethereal solution of oleoresin was extracted with two 200-ml portions of 2% caustic soda solution. Acidification of the aqueous alkaline solution to pH 2 with hydrochloric acid and its extraction with diethyl ether gave 10.61 g of residual acids. The yield of the neutral fraction was 13.00 g.

A sample of the neutral fraction (2.56 g) was dissolved with heating in 7 ml of hexane and the solution was left overnight in the refrigerator. The wax that deposited [0.56 g; IR spectrum, cm^{-1}: 1720 (C=O), 3620 (OH)] was filtered off and the mother liquor was chromatographed on silica gel giving successively 1.09 g of combined hydrocarbons, 0.20 g of a fraction of carbonyl compounds, 0.20 g of an intermediate fraction from which crystals of nonacosan-10-ol with mp 83-84°C (according to the literature [8] mp 83-84°C) deposited, 0.22 g of a fraction of monohydric polar alcohols, and 0.27 g of a fraction of polar compounds.

Isolation of the Esters (II), (III), and (IV). The combined acids isolated by extraction with caustic soda solution were methylated with diazomethane, and a sample of the esters obtained (3.25 g) was chromatographed on silica gel. The successive elution took place of 1.56 g of a mixture of the methyl esters of resin fatty acids (eluent: 3% of diethyl ether + 97% of petroleum ether), 0.80 g of the ester (I) (oil with n_D^{18} 1.5140, $[\alpha]_D^{19}$ + 46° (c 18.3); according to the literature [4]: $[\alpha]_D$ + 40° (c 3.65)), 0.29 g of the ester (II) and 0.30 g of the ester (III) in the form of crystals from a mixture of petroleum ether and diethyl ether, mp 39-40°C, $[\alpha]_D^{20}$ + 36° (c 13.0) (according to the literature [6]: mp 39-40°C).

Methyl 3-Ketoanticopalate (II). This was obtained in the form of an oil with n_D^{18} 1.5218, $[\alpha]_D^{20}$ + 34.4° (c 5.24). IR spectrum (in carbon tetrachloride), cm^{-1}: 900, 1650, 3087 (C=CH$_2$); 1150, 1220, 1230 (C–O); 1720 (broad, C=O). PMR spectrum (in carbon tetrachloride, 60 MHz, ppm): 0.80, 0.91, 0.97 (3 H each, singlets, tertiary methyl groups at C-4 and C-10); 2.05, 3.56 (3 H each, singlets, Me_{13} and COO<u>Me</u>, respectively); 4.53, 4.85 (1 H each, narrow multiplets C=CH$_2$); 5.50 (1 H, narrow multiplet, H_{14}).

Synthesis of the Ester (II). To a stirred solution of 0.20 g of the ester (I) in 20 ml of acetone was added 0.5 ml of Jones reagent, and the mixture was left at room temperature for 0.5 h. After the usual working up, 0.15 g was obtained of the ester (II) with $[\alpha]_D^{20}$ + 34.8° (c 5.17); its IR and PMR spectra coincided with those of a sample isolated from an extract of the needles under investigation.

Isolation of the Components of the Strong-Acid Fraction and of Methyl Sandaracopimarate. The strong-acid fraction (2.73 g) was methylated with an excess of an ethereal solution of diazomethane, and subsequent chromatography of the product on silica gel yielded 2.3 g of methyl benzoate (eluent: petroleum ether containing 5% of diethyl ether) and 0.1 g of the dimethyl ester of 19-O-succinylagatholic acid (eluent: petroleum ether containing 15% of diethyl ether) in the form of an oil with $[\alpha]_D^{20}$ + 33° (c 4.50); according to the literature [6]: $[\alpha]_D$ + 27° (c 3.4).

The fraction of methyl esters of the resin and fatty acids (1.56 g) obtained as described above was chromatographed on silica gel impregnated with 5% of silver nitrate. Petroleum ether containing 0.5% of diethyl ether eluted a mixture of fatty acid methyl esters (0.15 g), petroleum ether containing 3% of diethyl ether eluted a mixture of the methyl esters of dehydroabietic, neoabietic, levopimaric, and abietic acids (PMR spectrum) (1.05 g), and petroleum ether containing 5% of diethyl ether eluted methyl sandaracopimarate (0.15 g), the PMR spectrum of which (60 MHz, in deuterochloroform) coincided with that given in the literature [11].

Sandaracopimaradien-3β-ol (IV). The fraction of polar monohydric alcohols (0.22 g) was acetylated with acetic anhydride in pyridine (20°C, 12 h). Chromatography of the product on silica gel yielded 0.16 g of alcohol acetates and 0.04 g of α-cadinol. Rechromatography of the acetates so obtained on silica gel impregnated with silver nitrate led to the isolation of 0.14 g of a chromatographably homogeneous product, the reduction of which with lithium tetrahydroaluminate in diethyl ether (20°C, 1 h) gave 0.12 g of a mixture of the alcohols (IV) and (V), mp 126-127°C (from hexane), $[\alpha]_D^{20}$ − 12.66° (c 13.42). This product was dissolved in 5 ml of pyridine, and, with stirring and cooling to 0°C, 0.2 g of chromium trioxide was added and the mixture was left at the same temperature for 12 h.

After the usual working up, the product was chromatographed on silica gel. A mixture of petroleum ether + 3% of diethyl ether eluted 0.02 g of sandaracopimarinal, petroleum ether containing 5% of diethyl ether eluted 0.03 g of 3-ketosandaracopimaradiene (mp 59-60°C; IR spectrum (in KBr): 1705 cm^{-1} (C=O); according to the literature [14]: mp 59-60°C); and petroleum ether containing 20% of diethyl ether eluted 0.05 g of sandaracopimaradien-3β-ol (IV) with mp 129-130°C (from hexane), $[\alpha]_D^{17}$ − 22.7° (c 3.52).

Sandaracopimaradiene-3β,18-diol. Acetylation of the fraction of polar compounds (0.22 g) with acetic anhydride in pyridine (20°C, 12 h) followed by chromatography on silica gel led to the isolation of 0.18 g of sandaracopimaradiene-3β,18-diol diacetate with mp 132-133°C (from a mixture of hexane and diethyl ether), $[\alpha]_D^{22}$ + 16.4° (c 13.4); according to the literature [14]: mp 131.5°C, $[\alpha]_D$ + 13.5°. Its reduction with lithium tetrahydroaluminate in diethyl ether (20°C, 1 h) gave sandaracopimaradiene-3β,18-diol with mp 152-153°C (from a mixture of hexane and diethyl ether); according to the literature [14]: mp 152-153°C. PMR spectrum (in deuterochloroform), ppm: 0.84, 0.89, 1.02 (3 H each, singlets, tertiary methyl groups at C-4, C-10, and C-13); and 3.66 (1 H, doublet of doublets, J = 8.5 and 5.5 Hz, H_3). The methylene protons at C-18 appeared in the form of an AB system with components at 3.28, 3.46, 3.61, and 3.77 ppm. The signals of the olefinic protons coincided with those for methyl sandaracopimarate [11].

SUMMARY

1. The composition of an ethereal extract of the needles of the Japanese stone pine has been investigated and in it 33 compounds have been identified, 10 of which have been isolated in the individual state.

2. Derivatives of anticopalic acid — 3β-hydroxy-, 3β-acetoxy-, and 3-ketoanticopalic acids — accumulate in the needles of the Japanese stone pine.

3. The needles of the Japanese stone pine contain in considerable amounts sandaracopimaradien-3β-ol and sandaracopimaradiene-3β,18-diol, and this is the first time that these have been detected in plants of the family *Pinaceae*.

LITERATURE CITED

1. E. G. Bobrov, "Timber-Forming Conifers of the USSR [in Russian], Leningrad (1978), p. 119.
2. D. F. Zinkel, TAPPI Conference Papers, Madison, Wisconsin (1977), p. 53.
3. N. T. Mirov, Composition of Gum Terpentines of Pines, US Department of Agriculture (1961), p. 27.
4. V. A. Raldugin, V. A. Khan, Zh. V. Dubovenko, V. A. Pentegova, Khim. Prir. Soedin., 299 (1976).

5. V. A. Raldugin, L. I. Demenkova, and V. A.Pentegova, Khim. Prir. Soedin., 345 (1978).
6. V. A. Raldugin and V. A. Pentegova, Khim. Prir. Soedin., 158 (1983).
7. V. A. Raldugin, V. A. Khan, Zh. V. Dubovenko, and V. A.Pentegova, Khim. Prir. Soedin., 609 (1976).
8. V. A. Raldugin, N. V. Zubtsova, L. M. Shmakova, L. I. Demenkova, S. A. Ivashin, and V. A. Pentegova, Khim. Prir. Soedin., 112 (1983).
9. V. A. Raldugin and V. A. Pentegova, Khim. Prir. Soedin., 125 (1984).
10. F. S. Santamour and D. F. Zinkel, in: 1976 Proceedings of the 23rd Northeastern Forest Tree Improvement Conference, New Brunswick, N.J., Aug. 4-7 (1975), p. 46.
11. D. F. Zinkel, L. C. Zank, and M. F. Wesolowski, Diterpene Resin Acids. A Compilation of Infrared, Mass, Nuclear Mangetic Resonance, Ultraviolet Spectra and Gas Chromatographic Retention Data, US Department of Agriculture, Madison, Wis., (1971), D. 38.
12. R. A. Franich, L. G. Wells, and P. T. Holland, Phytochemistry, 17, 1617 (1978).
13. H. A. Candy, J. M. Pakshong, and K. H. Pegel, J. Chem. Soc., C, 2536 (1970).
14. R. A. Laidlaw, and J. W. W. Morgan, J. Chem. Soc., 644 (1963).
15. B. R. Thomas, Acta, Chem. Scand., 20, 1074 (1966).

STRUCTURE OF PSOLUSOSIDE A — THE MAIN TRITERPENE GLYCOSIDE FROM THE HOLOTHURIAN *Psolus fabricii*

V. I. Kalinin, A. I. Kalinovskii, and V. A. Stonik

UDC 547.996:593.96

The structure of psolusoside A — the main component of the glycosidic fraction from the holothurian *Psolus fabricii* Duben et Koren — has been determined as 3β-O-[O-(3-O-methyl-6-O-sulfato-β-D-glucopyranosyl)-(1 → 3)-O-(6-O-sulfato-β-D-glucopyranosyl)-(1 → 4)-O-β-D-quinovopyranosyl-(1 → 2)-β-D-xylopyranosyloxy]holosta-9(11),25-dien-16-one.

Continuing a chemical study of holothurians of the sublittoral of the island of Onekotan [1, 2] we have established the complete structure of psolusoside A — the main component of the glycosidic fraction from the holothurian *Psolus fabricii*. We had shown previously that this glycoside was a tetraoside of 3β-hydroxyholosta-9(11),25-dien-16-one, and tis monosaccharide residues were determined as those of D-xylose, D-quinovose, 3-O-methyl-D-glucose, and D-glucose [1]. We have now completed the interpretation of the structures of the carbohydrate chain of psolusoside A (I). (Formula, top, following page.)

The result of the solvolytic cleavage of psolusoside A with a mixture of pyridine and dioxane (1:1) indicated the presence of sulfate groups in the glycoside [3]. This solvolysis gave the desulfated derivative (II).

The Hakomori methylation of (II) [4] followed by methanolysis and acetylation led to the formation of methyl 2-O-acetyl-3,4-di-O-methyl-α- and -β-xylopyranosides, methyl 4-O-acetyl-2,3-di-O-methyl-α- and β-quinovopyranosides, methyl 3-O-acetyl-2,4,6-tri-O-methyl-α- and -β-glucopyranosides, and methyl 2,3,4,6-tetra-O-methyl-α- and -β-glucopyranosides, which were identified by GLC and chromato-mass spectroscopy. The results obtained showed that the carbohydrate chain was unbranched, and the terminal position was occupied by a 3-O-methylglucose residue.

On periodate oxidation of glycoside (I) followed by acid hydrolysis, the xylose and quinovose residues were destroyed, which indicated the absence of sulfate groups attached to these monosaccharide residues. The number and positions of the sulfate groups were established by comparing the ^{13}C NMR spectra of the desulfated derivative (II) and the glycoside (I) (Table 1).

Pacific Ocean Institute of Bioorganic Chemistry, Far Eastern Scientific Center, Academy of Sciences of the USSR, Vladivostok. Translated from Khimiya Prirodnykh Soedinenii, No. 2, pp. 212-217, March-April, 1985. Original article submitted April 25, 1984.

0009-3130/85/2102-0197$09.50