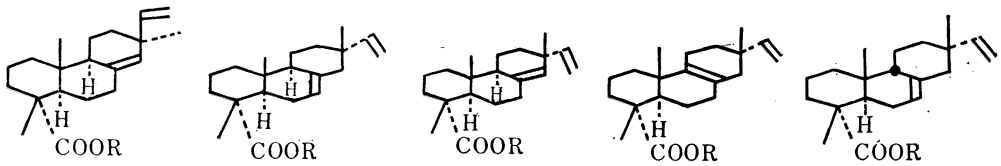
UDC 547.91

On Cryptopimaric Acid and Neoisopimaric Acid*1

The recent chemistry of the natural pimaric acid type compounds, such as pimaric acid (Ia),[1] m.p. 218~219°,[1a] $[\alpha]_D$ +73.3°(EtOH),[1a] isopimaric acid[1] (IIa) (miropinic acid[2]), m.p. 162~164°[1a] $[\alpha]_D$ ±0°(EtOH),[1a] sandaracopimaric acid[1f,1g,3] (IIIa), m.p. 171°,[3f] $[\alpha]_D$ −20°(EtOH),[3f] cryptopimaric acid,*2[1e,1f,3f,3g,4] m.p. 159~161°[4a] $[\alpha]_D^{17.5°}$ −18.99°(EtOH)[4a] and neoisopimaric acid (Ukita's acid),*2[1d,1f,3e,5] m.p. 162.5~165°,[5b] $[\alpha]_D^{24}$ −8.9° (Me_2CO-$CHCl_3$, 1:1),[5b] had been investigated in detail, but still remained some problems.



Ia R=H b R=Me IIa R=H b R=Me IIIa R=H b R=Me IVa R=H b R=Me Va R=H b R=Me

Especially, cryptopimaric acid and also neoisopimaric acid were now become the subjects of the discussion in the several reports. The former acid from the resin of *Cryptomeria japonica* D. Don (Sugi) was originally reported by S. Keimatsu *et al.*[4] in 1937. Recently Erdtman *et al.*[3f] succeeded in the separation of sandaracopimaric acid from cryptopimaric acid and also it was supported by the other studies using the physical methods.[1e] Independently ApSimon *et al.*[3g] claimed that the acid was a mixture of predominant isopimaric acid and/or neoisopimaric acid (Ukita's acid) with smaller amount of sandaracopimaric acid.

On the other hand, the latter, neoisopimaric acid (Ukita's acid), was firstly isolated from the fruits of *Juniperus rigida* Sieb. et Zucc. (Nezu) and *J. conferta* Parl. by Ukita *et al.*[5b] in 1952. While the same authors soon assigned[5c] (IVa) for its structure, Edwards

*1 The communication will be published in detail as Diterpenoids (III).

*2 Both acids, cryptopimaric acid and neoisopimaric acid, which had respectively the same melting point shown in the original references,[4a,5b] were used in these experiments.

1) *a*) J. Simonsen, D.H.R. Barton : The Terpenes III, 447, 457 (1952). *b*) B. Green, A. Harris, W.B. Whalley : J. Chem. Soc., **1958**, 4715; Chem. & Ind.(London), **1958**, 1084. *c*) E. Wenkert, J.W. Chamberlin : J. Am. Chem. Soc., **80**, 2912 (1958); **81**, 688 (1959). *d*) O.E. Edwards, R. Howe : Chem. & Ind.(London), **1958**, 629; **1959**, 537; Can. J. Chem., **37**, 760 (1959). *e*) H.H. Bruun, R. Ryhage, E. Stenhagen : Acta. Chem. Scand., **12**, 789 (1958); H.H. Bruun, I. Fishmeister, E. Stenhagen : *Ibid.*, **13**, 379 (1959); H.H. Bruun, E. Stenhagen : *Ibid.*, **13**, 832 (1959). *f*) A.K. Bose, W.A. Struck : Chem. & Ind.(London), **1959**, 1628; A.K. Bose : *Ibid.*, **1960**, 1104. *g*) R.E. Ireland, P.W. Schiess : Tetrahedron Letters, **1960**, No. 25, 37. *h*) J.C.W. Chien : J. Am. Chem. Soc., **82**, 4762 (1960). *i*) G.W.A. Milne, H. Smith : Chem. & Ind.(London), **1961**, 1307. *j*) R.F. Church, R.E. Ireland : Tetrahedron Letters, **1961**, No. 14, 493. *k*) W. Antkowiak, J.W. ApSimon, O.E. Edwards : J. Org. Chem., **27**, 1930 (1962). *l*) R.E. Ireland, J. Newbould : *Ibid.*, **27**, 1931 (1962).

2) A. Brossi, O. Jeger : Helv. Chem. Acta., **33**, 722 (1950).

3) *a*) T.H. Henry : J. Chem. Soc., **79**, 1144 (1901). *b*) A. Tschirch, M. Wolff : Archiv. Pharm., **244**, 684 (1906). *c*) F. Balaš, J. Brazák : Coll. Czech. Chem., **1**, 352 (1929); **2**, 424 (1930). *d*) F. Petrů, V. Galik : Coll. Czech. Chem., **18**, 717 (1953). *e*) O.E. Edwards, A. Nicolson, M.N. Rodger: Can. J. Chem., 38, 663 (1960). *f*) V.P. Arya, C. Enzell, H. Erdtman, R. Ryhage : Acta. Chem. Scand., **15**, 682 (1961). *g*) J.W. ApSimon, B. Green, W.B. Whalley : J. Chem. Soc., **1961**, 752.

4) *a*) S. Keimatsu, T. Ishiguro, G. Fukui : Yakugaku Zasshi, **57**, 69 (1937). *b*) T. Kondo, H. Imamura, M. Suda : Bull. Agri. Chem. Soc. Japan, **23**, 233 (1959).

5) *a*) T. Ukita, R. Matsuda : Yakugaku Zasshi, **71**, 1050 (1951). *b*) T. Ukita, T. Tsumita : *Ibid.*, **72**, 1324 (1952). *c*) T. Ukita, T. Tsumita, N. Utsugi : This Bulletin, 3, 441 (1955).

et al.[1d] proposed a new formula (Va)[1f] against Ukita's determination.

For the purpose of the reinvestigation of these problems concerning above mentioned two acids, gas chromatography, using a column packed with 1% QF-1-0065 on Gas-Chrom. P,*5 of the methyl esters of the natural pimaric acid series*3 including $\Delta^{13,14}$-

TABLE I. Relative Retention Time[a,b] of the Methyl Ester of the Pimaric Acid

Compound	Time	Ratio of area*4
Methyl $\Delta^{13,14}$-sandaracopimarate (IVb)	1.00[c]	
Methyl pimarate (Ib)	1.12	
Methyl sandaracopimarate (IIIb)	1.21	
Methyl isopimarate (IIb)	1.45	
Methyl cryptopimarate	1.20 & 1.44	4:3
Methyl neoisopimarate (Methyl ester of Ukita's acid)	1.21 & 1.45	3:1

a) Shimadzu GC-1B (hydrogen flame ionization detector), 1% QF-1-0065 (fluoroalkyl silicon polymer) phase on Gas-Chrom. P (100–120 mesh),*5 225 cm., 4 mm., i. d. column, pressure. 1.8 kg/cm^2, column temp. 165°, flash temp. 220°, cell temp. 210°, sensitivity 1000 MΩ, range 1.6.

b) The order of the increasing of the retention time was exactly similar to that of pimaradiene analogs.[1j]

c) Retention time, 5.65 min.

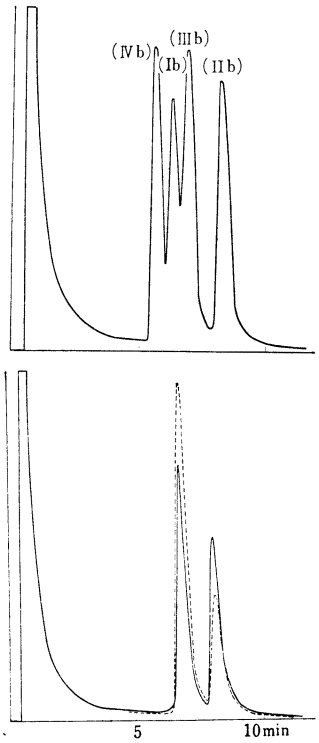


Fig. 1. Gas Chromatogram of a mixture of four methyl esters

Fig. 2. Gas chromatographic Behavior :
—— Methyl cryptopimarate
····· Methyl neoisopimarate
(Methyl ester of Ukita's acid)

*3 The samples of the pure acids were donated from the following peoples and so the authors gratefully acknowledge to them. Pimaric acid, m.p. 209~210°, from Prof. E. Wenkert, Bloomington, and Dr. R.V. Lawrence, Olustee, sandaracopimaric acid, m.p. 169~170°, $[\alpha]_D^{28.5}$ −24.3°(c=0.4, EtOH), $\Delta^{13,14}$-sandaracopimaric acid, m.p. 104~105°, from Dr. O.E. Edwards, Ottawa, isopimaric acid, m.p. 161~163°, from Prof. T. Tsumita, Tokyo, cryptopimaric acid, m.p. 159~161°, $[\alpha]_D^{29}$ −16.1° (c=0.43, EtOH), from Dr. H. Imamura, Tokyo and also neoisopimaric acid (Ukita's acid), m.p. 162~165°, $[\alpha]_D^{29.3}$ −21.6°(c=0.73, EtOH), was isolated by the use of the Ukita's method from the crude extract of the fruits of *Juniperus rigida Sieb. et Zucc.*, which was donated from Prof. K. Tsuda, Tokyo. (Physical constants were measured by us.)

*4 The measurements of these areas were calculated from the conventional method of multipling peak height by width at half height. cf. E.O.A. Haahti, W.J.A. Vanden Heuvel, E.C. Horning: Ann. Biochem., 2, 182 (1961).

*5 Thanks are due to Dr. E.C. Horning, Baylor Univ., for providing of the packing.

sandaracopimaric acid (IVa),*3,1d) which was easily rearranged from sandaracopimaric acid (IIIa), isopimaric acid (IIa) and also neoisopimaric acid (Ukita's acid), was carried out and the obtained results are shown in Table I, Fig. 1 and 2.

Based on these observations, we would like to propose herein the following conclusion that cryptopimaric acid and neoisopimaric acid are mixtures consisting mainly of sandaracopimaric acid (IIIa), together with isopimaric acid (IIa) (the ratio of the areas of the peaks in the gas chromatogram are 4:3 for cryptopimaric acid and 3:1 for neoisopimaric acid respectively*4) and therefore pimaric acid (Ia), isopimaric acid (IIa) and sandaracopimaric acid (III a) are only known as the natural pimaric acid type diterpene in pure state.

The gas chromatographic results were also supported by the facts that sandaracopimaric, cryptopimaric and neoisopimaric acid showed the undepressed mixed melting points with each other and had the nearly superimposable infrared spectra.

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A Novel Phosphorylating Agent, P-Diphenyl, P′-Morpholinopyrophosphorochloridate

Recently we have reported[1] the synthesis of morpholinophosphorodichloridate (I) and its use in the synthesis of nucleoside 5′-mono- and -polyphosphates. Because of the relatively weak phosphorylating power of this reagent, the first step of the phosphorylation could not exceed 50~60% extent. This fact would lead to the undesired side reaction, which affected greatly the purification procedure of the products. In order to increase the reactivity of the reagent and to avoid bifunctional reaction, P-diphenyl, P′-morpholinopyrophosphorochloridate (II) was synthesized and tested for the phosphorylation of nucleosides.

The reagent (II) was synthesized from diphenylphosphate (1 mmole) and morpholinophosphorodichloridate (1 mmole) in dioxane solution in the presence of 2,6-lutidine (2 mmole) as acid acceptor. After 15 minutes, reagent (II) was used *in situ* for the phosphorylation of 2′,3′-O-isopropylideneadenosine (IIIa) (0.5 mmole). 40 hours' reaction at 20° showed quantitative conversion of (IIIa) to 5′-(morpholino)phosphorochloridate (IVa) (Migratory distance 6.2 cm, R_{AMP} 0.58)[2] which was proved by paper electrophoresis (0.05*M* triethylammonium bicarbonate, pH 7.5,[3] 20 volt/cm. 1 hour[4]).

(IVa) was hydrolyzed with water (at pH 2.0, 70° for 30 minutes) and extracted with chloroform to remove diphenyl phosphate. After adjustment to pH 8.5 with lithium hydroxide, mixture was extracted again with ether to remove amines. Upon addition

1) M. Ikehara, E. Ohtsuka: This Bulletin, **10**, 536, 539 (1962).
2) This value was identical to that of AMP-morpholidate, which showed the hydrolysis of chloridate residue during electrophoresis.
3) J.G. Moffatt, H.G. Khorana: J. Am. Chem. Soc., **83**, 639 (1961).
4) This condition was used throughout present communication.