Summary

12-Methyl-1,2,3,4,6,7,12,12b-octahydro-2,6-methanoindolo[2,3-a]quinolizine (I), which represents the fundamental skeleton of sarpagine type indole alkaloids, was synthesized. Thus methyl ester (X) hydrochloride of N-benzyl-1-methyltryptophan was treated with methyl 3-formylpropionate to form tetrahydro-β-carboline derivative (XV), from which 6,10-imino-5H-cyclooct[b]indole derivative (XVII) was prepared by Dieckmann cyclization with sodium hydride under a certain working condition. This was condensed with methyl bromoacetate followed by ketone fission to yield ketoester (XX), from which N-benzyl group was reductively removed and then converted to N-methyl derivative (XXIII). The ethylene thioketal (XXV) of the latter was desulfurized as usual and the product was reduced by lithium aluminum hydride to furnish the corresponding alcohol (XXVII), which was tosylated with tosyl chloride in pyridine in the cold. Spontaneous quaternization of the tosylate took place forming the cyclized quaternary base (XXVIII), the chloride salt of which was submitted to pyrolysis reaction to produced the desired base (I).

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More than dozen pigments have been isolated from several strains of Penicillium islandicum Sopp grown on the Czapek–Dox medium.1) Erythroskynine which was first isolated and studied by Howard and Raistrick9) is unique among them as it contains nitrogen in its molecule. The present study has been carried out using laboratory cultivation of P. islandicum Sopp S-strain which was isolated from molded rice grains. The S-strain produces no anthraquinone pigments as similar as LSHTM BB 233 strain.2) A minor pigment accompanied by erythroskynine was removed by CaHPO4–column chromatography, using hexane–acetone (10:1) as the solvent, and the pure erythroskynine, orange red crystals, m.p. 130–133°, (α)D +46.9°, thus obtained gave a molecular formula, C24H18O6N, which was revised from the formula, C24H18O4N, proposed by the earlier workers.3)

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The purity of erythroskyrine was revealed by thin-layer chromatography to give a single spot whose identity with the main spot given by Professor Raistrick's sample of erythroskyrine was established. Erythroskyrine possesses an alcoholic hydroxyl which is proved by the formation of monoacetate whose infrared spectrum shows $\nu_{C-O}$ at 1740 cm$^{-1}$ and $\nu_{C-O}$ at 1250 cm$^{-1}$ (alcoholic acetate). The iodine–brown ferric reaction of erythroskyrine is also given by its monoacetate.

The presence of a polyene system in erythroskyrine which is suggested by a blue–violet color reaction with conc. H$_2$SO$_4$ is also revealed by the infrared spectrum absorptions at 1584, 1564, 1550, and 1010 cm$^{-1}$ for –CH–CH–, as well as by the nuclear magnetic resonance signals at $\tau$: 3.7–2.9 (10H) (in CDCl$_3$).

On catalytic hydrogenation in ethanol using platinum oxide as a catalyst, erythroskyrine absorbed 5 moles of hydrogen to afford a faint yellowish liquid, decahydro-erythroskyrine, which could not be purified owing to its instability.

The infrared absorption bands and the nuclear magnetic resonance signals of polyene system in the original erythroskyrine disappeared in the decahydro derivative, which gave strong infrared absorption at 2800–3000 cm$^{-1}$ region and nuclear magnetic resonance signals corresponding to 20H at $\tau$: 8.5 region.

Ozonization of erythroskyrine followed by hydrolysis with 2NH$_2$SO$_4$ afforded $\lambda(+)$-N-methylvaline (II) (DNQ-derivative, m.p. 194$^\circ$, [\alpha]$_D^{10}$ +441$^\circ$(in CHCl$_3$)), while by the same treatment decahydroerythroskyrine gave $\lambda(+)$N-methylvaline and decane dicarboxylic acid (III), m.p. 126.5$^\circ$. The $\beta$-triketone ring system in decahydroerythroskyrine is suggested by the enol ferric reaction and the formation of a green copper complex salt as well as by the infrared absorptions at 1710 (sh), 1690, 1635 (sh), and 1615 cm$^{-1}$ (in CHCl$_3$), and the ultraviolet absorptions at 225 m$_\mu$ (log $\varepsilon$ 3.86) and 284 m$_\mu$ (log $\varepsilon$ 4.04) (in EtOH).

The $\beta$-triketonic ring system in which the N-methylvaline moiety is involved as a lactam linkage has been established by the close resemblance of the ultraviolet and infrared spectra of decahydro derivative with those of tenuazonic acid (IV) which was obtained from Alternaria tenius Auct., and structurally established by Stickings. 3)

The isopropyl grouping and N–methyl lactam system in erythroskyrine were revealed by the nuclear magnetic resonance signals at $\tau$: 9.05 (3H) doublet (J = 7 c.p.s.), $\tau$: 8.90 (3H) doublet (J = 7 c.p.s.) for CH$_3$–CH$_2$–CH$_2$– system and $\tau$: 7.02 (3H), singlet for –CO–N–CH$_3$ system.

Decahydro-erythroskyrine was methylated with methyl iodide and potassium carbonate in acetone, and the product was treated subsequently with lithium aluminum hydride in absolute ether to afford a neutral compound, m.p. 64$^\circ$, a liquid amine and a crystalline amine, C$_{17}$H$_{26}$O$_3$N, m.p. 108$^\circ$, [\alpha]$_D^{10}$ +69$^\circ$(in CHCl$_3$), whose N–CH$_3$ signal in the nuclear magnetic resonance spectrum appeared at $\tau$: 7.72 which was shifted from the corresponding signal of erythroskyrine at $\tau$: 7.02 (in CDCl$_3$).

This indicated the conversion of a R–N–CO– system in erythroskyrine into a

\[ \text{Me} \]
\[ \text{R–N–CH$_3$– in the crystalline amine.} \]

The introduction of a C–CH$_3$ group by the methyl-

Me

ation of decahydroerythroskyrine was shown by the singlet CH$_3$ signal at $\tau$: 9.07 in the nuclear magnetic resonance spectrum and the negative ferric reaction of the crystalline amine.

According to the nuclear magnetic resonance spectrum, the neutral product, C$_{15}$H$_{24}$O$_4$, m.p. 64$^\circ$, [\alpha]$_D^{10}$ +31.1$^\circ$(in CHCl$_3$), seemed to retain the main part of the structure

of the original erythroskrynine, though it must lose the triketonic ring system probably by thermal decomposition during the process of methylation, since it showed neither nuclear magnetic resonance signals for tertiary C-Me and N-Me nor infrared absorption of carbonyl.

The presence of two hydroxyls, free and hydrogen bonded, respectively, in the neutral product was proved by the infrared spectrum giving doublet bands at 3632 and 3530 cm⁻¹ (in CCl₄ using CaF₂ prism).

The neutral compound gave diacetate, C₃₅H₃₅O₆, m.p. 29⁰, which showed no hydroxyl band in the infrared spectrum. On oxidation with the Kiliani reagent, the neutral compound afforded a monohydroxylic acid which was characterised by its monoacetate whose infrared spectrum (in CHCl₃) indicated the presence of a carboxyl group (1720 cm⁻¹) and an alcoholic acetate (1740 cm⁻¹). On oxidation with chromic acid at room temperature, the neutral compound gave decane dicarboxylic acid.

According to these results, it has been suggested that one of the hydroxyls of the neutral compound is a primary and other is in such a position that oxidation is hindered. The analytical results of the neutral compound allocated a bicyclic system involving two ethereal oxygen atoms, one of which is hydrogen-bonded with a hydroxyl. The doublet signal centered on ν : 8.73 of erythroskrynine and the neutral compound indicated the presence of –O–CH–CH₃ system.

Thus a structure (V) has been proposed for the neutral compound. The infrared absorption (in CHCl₃) at 3600 cm⁻¹ (free OH) 3535 (bonded OH),⁴ as well as the nuclear magnetic resonance spectral data of the model compound, dianhydrrosorbitol⁵ (Ⅵ), m.p. 61~63⁰, also supported the formula (V).

On treatment of diacetate of the neutral compound (Ⅵ) with boron trifluoride–ether complex in acetic anhydride⁶ followed by decacetylation with lithium aluminum hydride, a tetraol, C₃₅H₃₅O₆, m.p. 108⁰, was afforded. The nuclear magnetic resonance spectrum of the tetraol tetraacetate indicated that one of the ethereal linkages of the original neutral compound was cleaved by boron fluoride and the other ethereal ring was remained unaffected.

\[
\text{HOOC–(CH₂)₉–COOH} \\
\text{HOOC–(CH₂)₉–COOH}
\]

As one mole of periodate was consumed to oxidise the tetraol, while the neutral componud was not oxidised, the hydroxyl newly formed by the cleavage of ethereal linkage must be located at the adjacent position of the original alcoholic hydroxyl. The mass-spectrum of the neutral compound showed a peak at m/e 101, which would correspond to the ionic fragment (Ⅲ). All the evidences mentioned above showed that the formula (Ⅰ) represents most reasonably the structure of erythroserine.

The evidence for the ring structure of C_{160}-C_{166} has been provided by the study of nuclear magnetic double resonance spectrum of erythroserine measured in deuterochloroform at 100 Mc.p.s. (Fig. 1), and the results of spin-decoupling is shown in Table I.

**Table I. Nuclear Magnetic Double Resonance Data of Erythroserine**

<table>
<thead>
<tr>
<th>Proton (Position)</th>
<th>Irradiated (τ)</th>
<th>Observed (τ)</th>
<th>Multiplicity change</th>
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<tbody>
<tr>
<td>25-H</td>
<td>5.75</td>
<td>8.73</td>
<td>d→s</td>
</tr>
<tr>
<td>26-H</td>
<td>8.73</td>
<td>7.75</td>
<td>(a)*</td>
</tr>
<tr>
<td>23-H</td>
<td>5.96</td>
<td>7.75</td>
<td>(b)*</td>
</tr>
<tr>
<td>20-H</td>
<td>6.21</td>
<td>7.75</td>
<td>(c)*</td>
</tr>
<tr>
<td>21-H</td>
<td>5.50</td>
<td>6.21</td>
<td>(d)*</td>
</tr>
<tr>
<td>6-H</td>
<td>8.37</td>
<td>6.47</td>
<td>d→s Long range coupling</td>
</tr>
</tbody>
</table>

s: singlet d: doublet t: triplet q: quartet
(-): not decoupled

All the protons of erythroserine have been assigned as shown in Table I together with those given in Table II.

**Table II.**

<table>
<thead>
<tr>
<th>τ</th>
<th>Position</th>
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<td>(d)</td>
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<tr>
<td>7.05</td>
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a) Disappeared when measured on addition of acid.

**Experimental**

**Isolation of Erythroserine**—The mycelium of Penicillium islandicum SoPP Strain-S obtained by the cultivation on Czapek-Dox solution for 3 weeks at 25° in dark was separated from the culture filtrate, rinsed with water and immersed in a mixture of aq. 5% HCl and benzene. The mixture was treated by the method of Raistrick and Howard. The crude extract (25 g.) obtained from the culture
(30 L.) was chromatographed on a column of CaHPO₄ using hexane-acetone (10:1) as the developing solvent. The crude pigments separated into four bands, from the bottom to the top: (i) Pale yellow, (ii) orange red, (iii) orange red (main band), and (iv) red. From the main orange red band, crude erythroskline was obtained by elution in 10–15% yield of the extract. Repeated crystallization from EtOH afforded, pure erythroskline, orange red micro-needles, m.p. 130–133°. Anal. Caled. for CaH₃O₃N: C, 68.55; H, 7.30; N, 3.08. mol. wt., 455. Found: C, 68.33; H, 7.10; N, 2.99. mol. wt., 455 (Rast). OCH₃: nil. C–CH₃ (Kuhn–Roth) 5.12%. UV λₑₘₜₚₘp (log ε): 409 (4.45), 260 (3.95). λₚₘₜₚₘₚₘp (log ε): 392 (4.78), 260 (4.14). IR max (in CHCl₃, 1500–1700 cm⁻¹ region): 1550, 1564, 1584, 1612, 1635, 1693.

The pure crystalline erythroskline was identified by paper chromatography (Toyo Roshi No. S3, developing with petr. benzene-acetone–H₂O (5:3:5.3 (upper layer)) with the main spot given by Professor Raistrick's sample. The purity of erythroskline was established by thin-layer chromatography using Silicagel-G treated with 0.5N oxalic acid as adsorbent and a mixture of petr. benzene-acetone–H₂O (5:3:5.3 (upper layer)) as solvent to give Rf 0.29. Erythroskline gives an iodine-brown ferric color reaction in ethanol solution, and dissolves in conc. H₂SO₄ to give a blue–violet color. It is decolorized in Br₂-atmosphere to form a faint yellow liquid.

Erythroskline is soluble in acetone, CHCl₃, MeOH, EtOH, benzene, AcOH, and pyridine, and less soluble in ether, hexane, and petr. ether.

**Erythroskline Monoacetate**—On acetylation with Ac₂O and AcONa, erythroskline afforded monoacetate, which was purified by CaHPO₄–chromatography developed with hexane-acetone (10:1), and by recrystallization from hexane-acetone to give orange red micro-needles, m.p. 123°. Anal. Caled. for CaH₃O₃N: C, 67.6; H, 7.07; N, 2.82. Found: C, 67.42; H, 7.07; N, 2.92. UV λₑₘₜₚₘp (log ε): 402 (4.65), 260 (4.14), 270 (inflection). IR νₑₘₜₚₘₚₘp cm⁻¹: 1740 (alcoholic acetate). NMR δₑₘₜₚₘp (CH₃): 7.75 (3H, -O-C-CH₃). The acetate gives an iodine–brown ferric reaction in ethanol solution.

**Reduction of Erythroskline**—Erythroskline (500 mg.) dissolved in EtOH (50 ml) was catalytically reduced at 29° using PtO₂ (300 mg.) as a catalyst to absorb 5 moles of H₂ for about 1 hr. On evaporation of the solvent in vacuo a faint yellowish liquid was obtained which could not be purified owing its instability.

**Copper Salt of Reduced Erythroskline**—Reduced erythroskline (250 mg.) dissolved in EtOH (5 ml) was titrated with aq. N NaOH to pH 7, then treated with a little excess of 0.1N copper acetate (14 ml). The dark green reaction mixture was extracted with CHCl₃ to give a green residue on evaporation in vacuo, which was dissolved in warm MeOH and diluted with warm water. On cooling, a crystalline green copper salt was separated, which, however, melted at room temperature.

**Ozonolysis of Erythroskline**—Ozonized O₃ was passed through a solution of erythroskline (1 g.) in AcOH (30 ml). After 20 hr., when the orange red color of solution was decolorized, 30% H₂O₂ (7.5 ml) was added. After standing overnight the excess of H₂O₂ was decomposed by the addition of Pd, and then heated about 50° on a bath. A pale yellow liquid which was obtained on evaporation of solvent was heated on a boiling bath for 8 hr. in 2N H₂SO₄ (30 ml). The brown colored reaction mixture was passed through an IR–4B ion exchange resin column and then through an IR–120 ion exchange resin column, from which amino acids fraction was eluted with 5% NH₃–H₂O.

**PFC**—Toyo Roshi No. 50: BuOH–AcOH–H₂O (4:1:5) (upper layer).

Rf: 0.55 (Main spot): Sprayed with p–NO₂–benzoic chloride–pyridine–red, 0.48 (Minor spot): Sprayed with ninhydrin–purple.

**Dinitrophenylation of Crude Amino Acids**—To a solution of crude amino acid (0.1 g.) in H₂O (5 ml) and NaHCO₃ (0.4 g.), an ethanolic solution of 2,4-dinitrofluorobenzene (0.4 g. in 10 ml) was added and stirred for 2 hr. in dark at room temperature. The reaction mixture was concentrated and extracted with ether to remove the excess of 2,4-dinitrofluorobenzene, and then acidified with 2N HCl. The precipitates thus formed were extracted with CHCl₃, and the solution was dried with Na₂SO₄, and then concentrated. From the mixture DNPE-amino acids were separated by chromatography on a column of Celite–545 buffered with 0.4M phosphate solution at pH 7.0 using CHCl₃ as the developing solvent. Dinitrophenol was removed as the first band and from the main band, a crude DNP-amino acid was eluted, which was purified by repeated recrystallization from CHCl₃–petr. ether to give orange–yellow leaflets, m.p. 192–193° (decomp.). Anal. Caled. for C₉H₈O₃N₃: C, 48.48; H, 5.09; N, 14.14. Found: C, 48.59; H, 5.11; N, 14.34. [α]D + 440.9° (c=0.615, CHCl₃). The DNP-amino acid was proved to be identical with 1[+]-DNPN-ethylvaline by a mixed fusion, a comparison of IR spectra and thin-layer chromatograms (Rf: 0.32; silica gel G; BuOH–AcOH–CHCl₃=1:1:5:97.5).

A minor DNP-amino acid was obtained in orange yellow microcrystals, m.p. 128–132°, by a repeated recrystallization from CHCl₃–ligroin. This DNP-amino acid was proved to be identical with l-DNP-valine by a mixed fusion and a comparison of IR spectra and thin–layer chromatograms (Rf: 0.56). Anal. Caled. for C₉H₈O₃N₃: C, 46.64; H, 4.63; N, 14.84. Found: C, 46.44; H, 5.01; N, 14.82.

**Ozonolysis of Reduced Erythroskline**—Ozonolysis of reduced erythroskline was carried out by the same way described for erythroskline. After refluxing with 2N H₂SO₄, amorphous precipitates
were filtered off and washed with H₂O. The filtrate was extracted with ether repeatedly to obtain a brown liquid accompanied by colorless crystalline substance which was mixed with the amorphous product and sublimed in vacuo. The crystalline sublimate was recrystallized from H₂O to yield micro-needles, m.p. 128°, which were identified as decane dichromic acid by a mixed fusion with the authentic sample. The hydrazide derivate of this acid was colorless micro-needles (from MeOH), m.p. 190°. *Anal. Calcd. for C₁₈H₂₄O₁₀N₂: C, 55.87; H, 10.14; N, 21.69. Found: C, 55.71; H, 10.19; N, 21.46.* The identity of this hydrazide with decane dichromic acid hydrazide was proved by a mixed melting point and IR spectra. l-DNP-N-Methylvaline was obtained from the aqueous layer, which was characterized as the DNP derivative which was purified by chromatography on phosphate-buffered Celite 545 column to afford orange yellow leaflets (from CHCL₃-petr. ether), m.p. 192~193° (decomp.). The DNP-amino acid was proved to be identical with l-DNP-N-methylvaline by a mixed fusion, and comparison of IR spectra and thin-layer chromatograms.

**Methylation of Reduced Erythrosyme**—Reduced erythrosyme (1 g.) was refluxed with MeI (5 ml), K₂CO₃ (2.5 g) in acetone (25 ml) for 7 hr., and the mixture was heated subsequently for 10 hr. after successive addition of MeI (3 ml) and acetone (5 ml). The reaction mixture was filtered and washed with acetone, and the filtrate was evaporated in vacuo to give a pale yellow liquid which showed no coloration with FeCl₃ in EtOH. Purification by high vacuum distillation (1/1000 mm. Hg, 240~250° (bath temp.)) was not successful owing to its instability.

**Reduction of Methylylated Reduced Erythrosyme with Lithium Aluminum Hydride**—The ethereal solution of methylated reduced erythrosyme (1 g. in ether 40 ml) was added with the ethereal suspension of LiAlH₄ in 100 ml. ether, and refluxed for 30 hr. After decomposing the excess of LiAlH₄ by a cautious addition of a few drops of H₂O, 150 ml. of H₂O was added to the ethereal solution. The upper layer was separated and extracted with 2N HCl. The ethereal solution was dried on anhydrous Na₂SO₄, and evaporated in vacuo to give a colorless waxy substance which was recrystallized from hexane to afford colorless micro-needles. *m.p. 64°. Yield : 170 mg. Anal. Calcd. for C₁₈H₂₄O₂: C, 88.75; H, 10.90: mol. wt., 514. Found: C, 68.83; H, 10.91; mol. wt., 314 (Mass-spectrometric). [α]D+ + 31.1° (c=0.548, CHCl₃). O-CH₃ : nil (micro-Zeisel method, and NMR).

The HCl-layer was alkalisised with 2N NaOH and extracted with ether to give an oily pale yellow residue on evaporation of the solvent, which was crystallized from acetone-H₂O to afford colorless needles, m.p. 109°. (Yield : 150 mg.). *Anal. Calcd. for C₁₈H₂₄O₂N₂: C, 69.04; H, 10.95; N, 2.98. mol. wt., 493.7. Found: C, 69.26; H, 10.89; N, 2.99. mol. wt., 462 (Rast). [α]D+ + 69.0° (c=0.771, CHCl₃). O-CH₃ : nil (micro-Zeisel method and NMR) N-CH₃ 1 (NMR). It gave an orange red color with Dragendorf's reagent, and a brown violet color with platinum iodide. From the mother liquor of recrystallized amine, a faint yellow oily amine was obtained, which could not be purified by vacuum distillation owing its instability (Yield : about 500 mg.). The same color reactions were given by this liquid amine as observed in crystalline amine. O-CH₃ : nil (micro-Zeisel method).

**Acetylation of Neutral Product (Diol)**—To a cold solution (6 ml.) of diol (200 mg.) in pyridine AcCl (3 ml.) was added under cooling, and the solution was allowed to stand overnight. The reaction mixture was poured in ice water. The precipitates formed were collected and recrystallized from MeOH-H₂O to give colorless needles, m.p. 29°. *Yield : 160 mg. Anal. Calcd. for C₁₈H₂₄O₂: C, 66.30; H, 9.61. Found: C, 66.21; H, 9.52. IR ν(CH₃)cm⁻¹: νC=O 1748 (O-COCCH₃), νC=C 1244; NMR δ(CH₃): 7.93, 7.89 (2 O-COCCH₃).

**Acetylation of Diacetyl Diol with Acetic Anhydride and Boron Fluoride**—To a cold Ac₂O soln. (3 ml.) of diacetyl of the neutral product (diol) (50 mg.) was added 3 drops of BF₃-ether complex under ice cooling, and the mixture was allowed to stand for 1 hr. at room temperature. The resulting faint brown reaction mixture was poured into ice water and after the excess of Ac₂O was decomposed, the reaction mixture was extracted with ether. From the ethereal solution, the solvent was removed in vacuo, to give a pale yellow oily residue which was refluxed in ether (30 ml.) with LiAlH₄ (500 mg.) for 5 hr. After decomposing the excess of LiAlH₄ by a cautious addition of a few drops of dil. H₂SO₄, 50 ml. of H₂O was added to the ethereal solution. The ethereal layer was dried over Na₂SO₄ and evaporated to dryness. The colorless precipitates separated in aqueous layer were filtered off, washed with H₂O and dried in vacuo. The both products were mixed and recrystallized from CHCl₃ to give colorless needles, m.p. 108°. *Yield : 30 mg. Anal. Calcd. for C₁₈H₂₄O₂: C, 65.02; H, 10.92. Found: C, 64.95; H, 10.80.

**Oxidation of Neutral Product (Diol) with Chromium Trioxide**—A solution of CrO₃ (60 mg.) in AcOH (1.4 ml.) and H₂O (0.1 ml.) was added dropwise to a solution of the neutral product (100 mg.) in AcOH (30 ml.). The mixture was stirred at room temperature for 22 hr. and then poured into ice-water. The precipitates thus formed was extracted with ether and washed with H₂O. The etheral solution was dried over Na₂SO₄ and evaporated in vacuo dryness to give a residue which showed an IR absorption band of COOH at 1720 cm⁻¹ (in CHCl₃). An excess of CH₃N₃ in ether was added to the ethereal solution of the acid and the mixture was allowed to stand for a few minutes. The residue obtained on evaporation showed an IR absorption band at 1740 cm⁻¹ (ester C=O, in CHCl₃). The ester was dissolved in EtOH and added with NH₄OH-H₂O. The mixture was heated on a boiling water-bath for 1 hr. On concentration
of the reaction mixture, colorless crystals separated out, which on recrystallization from MeOH afforded colorless micro-needles. The product was proved to be identical with decane dicarboxylic acid hydrazide by a mixed fusion and a comparison of IR spectra with the authentic sample.

Dianhydroisorbitol—Dianhydroisorbitol was prepared by refluxing sorbitol in conc. HCl for 24 hr., and the reaction mixture was treated by the method of Montgomery, et al. m.p. 60°. Anal. Calcd. for C₄H₁₀O₄: C, 49.31; H, 6.90. Found: C, 48.93; H, 7.07. IR cm⁻¹ νOH 3600 (free, OH), 3535 (bonded OH). (in CHCl₃ dilute solution: CaF₂ prism.)

Estimation of the Consumption of Sodium Periodate during Oxidation of Tetroal and Diol—To a solution of tetroal or diol (1~2 × 10⁻⁴ M) in MeOH–H₂O (3:2) or AcOH–H₂O, 0.01M NaI₀₃ solution (3 ml) was added and the mixture was allowed to stand in dark at 25°. The solution was added with saturated NaHCO₃ solution (5 ml), 0.0423 M standard solution of Na₂S₂O₃ and 20% KI solution. After standing for 30 sec., the solution was titrated with I₂ solution (0.05 M) using soluble starch solution as the indicator.

<table>
<thead>
<tr>
<th>Material</th>
<th>Tetraol</th>
<th>Diol</th>
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<tbody>
<tr>
<td>Solvent</td>
<td>MeOH–H₂O</td>
<td>MeOH–H₂O</td>
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<tr>
<td>Time (hr.)</td>
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<td>3.5</td>
</tr>
<tr>
<td>NaI₀₃ (moles)</td>
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Oxidation of Diol with the Kiliani Reagent—To a solution of diol (100 mg.) in acetone (40 ml.), the Kiliani reagent (0.6 ml.) was added dropwise under stirring at room temperature. After 1/2 hr., the reaction mixture was poured in H₂O (50 ml.) and then extracted with ether. Ether was distilled off to give an oily residue which was examined by paper chromatography. (Toyo Roshi No. 50. Solvent : PrOH-conc. NH₄OH (70:30 v/v). Developing agent : Universal indicator). The Rf of oxidative product (0.92) differed from the Rf of decane dicarboxylic acid (0.87). The crude product dissolved in ether was extracted with 5% aq. NaHCO₃. Ethereal layer was washed with H₂O, dried over Na₂SO₄ and evaporated. The IR spectrum of the residue was examined to give bands at 3480 cm⁻¹ (OH) and 1727 cm⁻¹ (C=O) (in CHCl₃). The alkaline solution was neutralized with dil. HCl and extracted with ether. The residue obtained by evaporation was acetylated with Ac₂O-pyridine at room temperature. The reaction mixture was poured in H₂O and extracted with ether. The solvent was removed to obtain residue which was examined by IR spectrum in CHCl₃. IR νCH₂ cm⁻¹ : 1720 (carboxylic acid), 1740 (alcoholic acetate).

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Summary

The structure of erythroskyrine, C₂₅H₃₉O₄N, m.p. 130~133°, [α]D +46.9°, an orange red pigment of Penicillium islandicum Sopp has been elucidated to be represented by the formula (I).

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