Summary

A new method for synthsis of β-carboline derivatives reported in the previous papers was now extended to include the preparation of tetrahydrobenzindoloquinolizine derivative (XIII). Thus, N-(3,4-dimethoxyphenethyl)-2-indoleacetamide was cyclized by Bischler-Napieralski-Perkin method to yield the corresponding 3,4-dihydroisoquinoline. The latter, after being dehydrogenated, was condensed with formaldehyde and diethylamine to give the gramine-type compound (V), from which benzindoloquinolizinium base (XII) was prepared as previously. Reduction of the latter to (XIII) was effected either catalytically or by means of sodium borohydride.

(Received January 30, 1960)

UDC 547.976 : 582.282.12

145. Shoji Shibata and Isao Kitagawa : Metabolic Products of Fungi. XVI.*[2] The Structures of Rubroskyrin and Luteoskyrin. (2).

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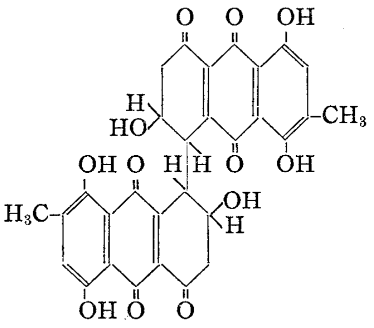
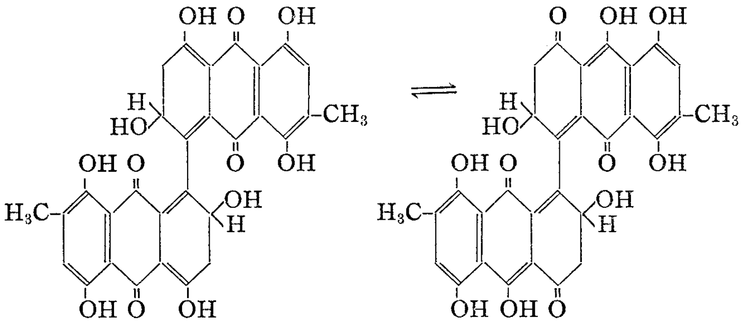
In a previous paper on the coloring matters of *Penicillium islandicum*,[1)] the structures of rubroskyrin (I) and luteoskyrin (II) were proposed and an isomerization reaction of rubroskyrin into luteoskyrin was described.



Rubroskyrin (I)



Luteoskyrin (II)

The properties of both pigments are summarized in Table I.

The present paper deals with the infrared spectral analysis of rubroskyrin and luteoskyrin, and provides some additional evidences for the presence of alcoholic hydroxyls in the molecules of both pigments.

The infrared stretching absorption band of carbonyl in rubroskyrin undergoes a shift towards higher frequency (1703 cm^{-1}) than that usually expected for a six-membered α,β-unsaturated ring ketone.

As the model compounds for rubroskyrin and luteoskyrin, 3,4-dihydro-1,9,10(2*H*)-anthracenetrione[2)] (VII), 3,4-dihydro-8-hydroxy-1,9,10(2*H*)-anthracenetrione (VIII), 1-hydroxy-

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*[1] Hongo, Tokyo (柴田承二, 北川 勲).

*[2] Part XV. S. Shibata, S. Natori, K. Fujita, I. Kitagawa, K. Watanabe : This Bulletin, **6**, 608(1958).

1) Part (1). S. Shibata, I. Kitagawa : *Ibid.*, **4**, 309(1956).

2) K. Zahn, H. Koch : Ber., **71**, 172(1938).

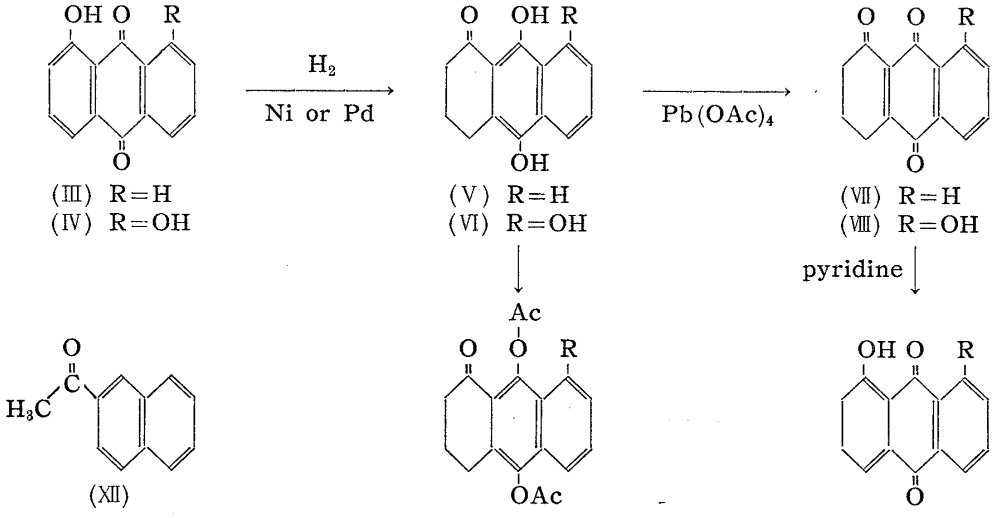
Table I.

	Rubroskyrin	Luteoskyrin
Color	brown-red	yellow
m.p.[a]	281°(decomp.)	281°(decomp.)
$[\alpha]_D$ (in Me_2CO)	levorotatory[b]	−880°
2*N* NaOH	green	orange yellow
conc. H_2SO_4	wine-red → blue	orange red ⟶ reddish purple
$Mg(OAc)_2$	+ (green)	− (yellow)
Photosensitivity	−	sensitive : ⟶ lumiluteoskyrin
Toxicity	−	toxic, causing liver cirrhosis
IR ν_{max}^{Nujol} cm^{-1}		
C=O	1703, 1623	1623
O-H	3559(w), 3341	3378

a) A mixed fusion of the two pigments showed a depression in m.p. of about 5°.

b) The deep light absorption of rubroskyrin solution prevented exact measurement of optical rotation.

2,3-dihydroanthraquinone (IX), and 1,8-dihydroxy-2,3-dihydroanthraquinone[3] (X) were prepared.



(III) R=H
(IV) R=OH

(V) R=H
(VI) R=OH

(VII) R=H
(VIII) R=OH

(XII)

(XI)

(IX) R=H (X) R=OH

The infrared absorption maxima of the compounds (V∼XI) are listed in Table II.

Table II. Infrared Absorption Maxima (cm^{-1}) (in Nujol)

Compound	(V)	(VI)	(VII)	(VIII)	(IX)	(X)	(XI)
C=O	1632	1631	1704 1667 1620	1710 1670 1634	1707 1625	1717 1623	1752 1689
O-H	3349	3324 3500	—	—	3444*	3294*	—

* The O-H absorption band must be attributed to the presence of tautomeric form of the compounds (IX) and (X).

It is noted that the compounds (VII) and (VIII) showed the C=O stretching absorptions in higher frequencies than in rubroskyrin, at 1704 and 1710 cm^{-1}, respectively.

The absorption spectra (ultraviolet and visible region) of the diacetate (XI) and 2′-acetonaphthone (XII), which showed almost superimposable curves (Fig. 1), gave an evidence for the validity of the structure (XI), proving the structure of (V) to be correct.

The compound (XI) showed the infrared absorption bands at 1752(acetyl), 1689(non-chelated C=O), and 1628(phenyl) cm^{-1}, among which the non-chelated C=O band appeared

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[3] S. Shibata, T. Murakami, M. Takido: This Bulletin, 4, 303(1956).

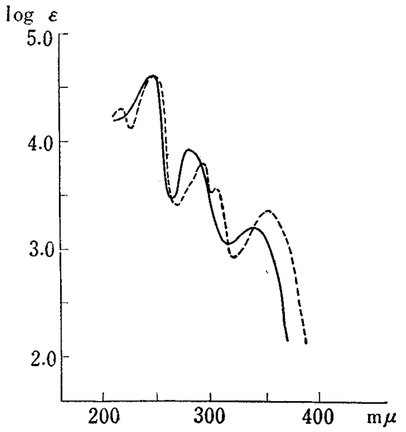
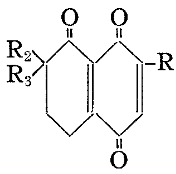


Fig. 1.

—— 2′-Acetonaphthone (XII)

------ 9,10-Diacetoxy-3,4-dihydro-1(2*H*)-anthracenone (XI) (in EtOH)

at a lower frequency than that in compounds (VII) and (VIII). This would indicate, as was also pointed out by Shoji[4] with 7,8-dihydro-1,4,5(6*H*)-naphthalenetrione, (XIII) and (XIV), that the neighboring quinone system causes a shift of absorption of C=O towards higher frequency.



(XIII) $R_1=R_2=CH_3$, $R_3=H$; ν_{max} 1712 cm^{-1} (C=O)

(XIV) $R_1=R_2=R_3=CH_3$; ν_{max} 1701 cm^{-1} (C=O)

As described in a previous paper, rubroskyrin was isomerized readily into luteoskyrin on being boiled in chlorobenzene in the presence of pyridine. It was found later that this isomerization of rubroskyrin also occurs (i) on boiling with dimethylaniline, (ii) on standing the pyridine solution for a few days at room temperature, and (iii) on heating the pyridine solution for a few minutes by which a most satisfactory result was obtained.

Luteoskyrin thus obtained showed $[\alpha]_D^{23.4}$ −857°±19° (c=0.107 in acetone), being identical with natural luteoskyrin. This type of isomerization was observed in the compounds (VII) and (VIII) giving (IX) and (X), respectively.

On acetylation with acetyl chloride in acetic acid, rubroskyrin yielded a diacetate, $C_{30}H_{20}O_{10}(OAc)_2$, m.p. 196° (decomp.); IR ν_{max}^{KBr} cm^{-1} : 1749 (alcoholic acetate C=O), 1708 (non-chelated C=O), 1620 (chelated C=O overlapped phenyl), and 3538 (crystal water). The ultraviolet and visible-region absorption spectra of the diacetate very closely resembled that of rubroskyrin (Fig. 2).

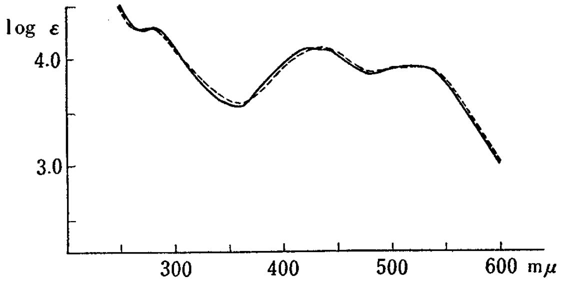


Fig. 2.

—— Rubroskyrin (I)

------ O-Diacetylrubroskyrin (XV) (in $CHCl_3$)

These facts indicated that the acetylation proceeded at the alcoholic hydroxyls of rubroskyrin, which are not conjugated with the quinone system. Consequently, the presence of alcoholic hydroxyls has now been confirmed at the 2- and 2′-positions[*3] in rubro-

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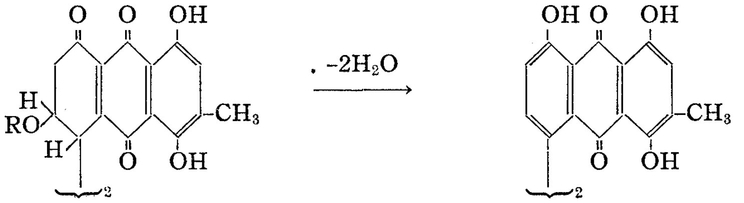
[*3] The presence of hydroxyls in the 2- and 2′-positions of rubroskyrin was suggested by the formation of catenarin on thermal decomposition.[1]

4) T. Shoji : Yakugaku Zasshi, **79**, 1034(1959).

skyrin molecule (I).

It would be noted that O-diacetylrubroskyrin was transformed readily into iridoskyrin1) on boiling its ethanolic solution for a few minutes, while this was not the case in chloroform solution. This must be the result of a *trans*-elimination of AcO at C-2 and H at C-3 or C-1 by the action of ethanol, suggesting an axial conformation of the alcoholic hydroxyl of rubroskyrin.



(XV) R=CH_3CO Iridoskyrin (XVI)
(I) R=H (Rubroskyrin)

After some unsuccessful attempts using usual acetylation agents, luteoskyrin yielded O-octaacetylluteoskyrin, $C_{30}H_{14}O_4(OAc)_8$, m.p. 285.5°(decomp.), by treatment with acetic anhydride and 60% perchloric acid. The presence of alcoholic hydroxyl in luteoskyrin was now established by the infrared absorption maxima (in dioxane) of the octaacetate at 1751 (alcoholic acetate C=O) and 1774 cm^{-1} (phenolic or enolic acetate C=O).

The C=O bands of alcoholic acetate in O-diacetylrubroskyrin and O-octaacetylluteoskyrin appear in higher frequencies than usually expected, but a similar example has been seen in O-acetylgeigerin (1751 cm^{-1} in $CHCl_3$).5)

Experimental

9,10-Dihydroxy-3,4-dihydro-1(2*H*)-anthracenone (V)—1-Hydroxyanthraquinone (11.2 g.) suspended in chlorobenzene (50 cc.) was catalytically hydrogenated over Raney Ni (0.5 g.) catalyst at 80~100°(starting at 25°) under a pressure of 75 kg./cm^2, unitl 2 moles of H_2 was absorbed in 8 hr. The reaction mixture was filtered, and the orange-brown crystalline powder which was obtained on evaporation was dissolved in benzene and chromatographed over $CaHPO_4$ column, when it separated into 2 bands. The starting material was recovered from the lower yellow band and the pale yellow upper band which showed a yellowish blue fluoresence under UV irradiation gave a yellow eluate from which orange crystalline precipitate (500 mg.) separated on concentration. The precipitate was purified by sublimation *in vacuo* (0.002 mm. Hg) at 130~140°, and recrystallization from AcOH to obtain yellow needles, m.p. 171°. *Anal.* Calcd. for $C_{14}H_{12}O_3$: C, 73.67; H, 5.30. Found : C, 73.66; H, 5.22.

9,10-Diacetoxy-3,4-dihydro-1(2*H*)-anthracenone (XI)—A mixture of 9,10-dihydroxy-3,4-dihydro-1(2*H*)-anthracenone (100 mg.), Ac_2O (1 cc.), and anhyd. AcONa (0.2 g.) was boiled for 30 min. The reaction mixture was poured into ice-water, when a white precipitate separated out and was recrystallized from EtOH to colorless needles, m.p. 216.5~218.5°. *Anal.* Calcd. for $C_{18}H_{16}O_5$: C, 69.22; H, 5.16. Found : C, 69.46; H, 5.12. UV $\lambda_{\max}^{\mathrm{EtOH}}$ mμ (log ε) : 216 (4.32), 248 (4.61), 294 (3.77), 355 (3.55). 2′-Acetonaphthone, UV $\lambda_{\max}^{\mathrm{EtOH}}$ mμ (log ε) : 246 (4.64), 337 (3.21), 282 (3.94).

3,4-Dihydro-1,9,10(2*H*)-anthracenetrione (VII)—9,10-Dihydroxy-3,4-dihydro-1(2*H*)-anthracenone (100 mg.) was suspended in AcOH (2 cc.) when the yellowish color of the mixture changed instantly into orange. The mixture was shaken vigorously for 5 min., when some slightly pink-colored leaflets separated. The mixture was carefully diluted with a small amount of distd. H_2O to complete the precipitation and the precipitate was collected by rapid filtration with suction. The crystals were washed successively with distd. H_2O and a small amount of MeOH, and recrystallized from chlorobenzene, m.p. 147.5~148.5°. *Anal.* Calcd. for $C_{14}H_{10}O_3$: C, 74.33; H, 4.46. Found : C, 74.05; H, 4.66.

From the mother liquor of recrystallization, the isomerized compound (IX) was obtained.

1-Hydroxy-2,3-dihydroanthraquinone (IX)—A yellow precipitate separated from the filtrate obtained after removal of leaflets (VII) on recrystallization after standing for a few days. It was recrystallized from chlorobenzene to yellow prisms, m.p. 201°(decomp.). *Anal.* Calcd. for $C_{14}H_{10}O_3$: C, 74.33; H, 4.46. Found : C, 74.17; H, 4.46.

8-Hydroxy-2,3-dihydro-1,9,10(2*H*)-anthracenetrione (VIII)—To the suspension of tetrahydrochrysazin[*4] (700 mg.) in AcOH (7 cc.) a suspension of $Pb(OAc)_4$ (1.4 g.) in AcOH (17.5 cc.) was added, when

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[*4] Prepared as given in Footnote (3).

5) D. H. R. Barton, J. E. P. Levisalles : J. Chem. Soc., **1958**, 4518

yellow color of the mixture changed to orange immediately. The reaction mixture was shaken at room temperature for 10 min. and formed a brownish orange clear solution. After removal of the remaining starting material, the filtrate was diluted with distd. H_2O to obtain a clear pink-brownish precipitate which was immediately collected by filtration and washed sucessively with H_2O and MeOH.[*5] Yield, 400 mg. Recrystallization from a small amount of dioxane (ca. 4 cc.) gave pink leaflets, m.p. 164~166°, which must be separated from the mother liquor as soon as possible to avoid isomerization into the compound (X).

The crystals of (Ⅷ) shows a brownish purple coloration in ethanolic solution of $Mg(OAc)_2$ which changes instantly into yellow by isomerization into the compound (X). In conc. H_2SO_4, the compound (Ⅷ) gave brownish purple color.

The purple coloration of the compound (Ⅷ) in 2*N* NaOH changed into orange by the addition of $Na_2S_2O_4$ and the original quinonic compound (Ⅷ) was not recovered on air oxidation but a nonquinonic compound (X) was obtained. *Anal.* Calcd. for $C_{14}H_{10}O_4$: C, 69.42; H, 4.16. Found[*6] : C, 69.63; H, 4.61.

O-Diacetylrubroskyrin (XV)—To a suspension of rubroskyrin (100 mg.) in glacial AcOH (20 cc.), AcCl (5 cc.) was added under ice-cooling and the mixture was occasionally shaken for 20 min. under cooling and another 10 min. at room temperature. The reaction mixture was then allowed to stand overnight and poured into ice-water (200 cc.) containing pyridine (1 cc.). The red precipitate formed was collected, washed with distd. H_2O, and purified by chromatography over $CaHPO_4$-column using benzene-Me_2CO (40:1) as the solvent. Three main bands were separated to form, from bottom to top, (a) pink, (b) yellow, and (c) red bands. From the pink band (a), red amorphous substance was recovered (m.p. 346~347°; yield, 24 mg.), which was acetylated with Ac_2O and AcONa to give O-hexaacetyliridoskyrin (m.p. and mixed m.p. 269~269.5°). From the yellow band (b), a small amount of orange powder was obtained, which was supposed to be a highly acetylated product but failed to purify. O-Diacetylrubroskyrin was separated from the band (c), which was recrystallized from Me_2CO-hexane mixture to form dark red crystals m.p. 193~196° (decomp.). Yield, 42 mg. *Anal.* Calcd. for $C_{30}H_{20}O_{10}(OAc)_2 \cdot H_2O$: C, 60.38; H, 4.42. Found : C, 60.35; H, 4.17.

Rf value on paper chromatogram developed with upper layer of Me_2CO-benzine-H_2O (5:5:3.5) 0.94, and upper layer of the same solvent combination (1:1:3) 0.56 (with tailing). UV $\lambda_{max}^{CHCl_3}$ mμ (log ε): 274 (4.31), 424 (4.12), 520 (3.95). Rubroskyrin : UV $\lambda_{max}^{CHCl_3}$ mμ (log ε) : 275 (4.31), 415 (4.10), 435 (4.10), 530 (3.97), 540 (3.97).

O-Diacetylrubroskyrin gave a green coloration with $Mg(OAc)_2$ in MeOH and its color changed into purple. On boiling with EtOH, O-diacetylrubroskyrin was converted into iridoskyrin (XVI) and, on allowing to stand for 2 days in EtOH at room temperature, 60% of the diacetate was transformed into iridoskyrin.

O-Octaacetylluteoskyrin—Luteoskyrin (300 mg.) was suspended in Ac_2O (5 cc.) and a mixture of 60% $HClO_4$ (1 drop) and Ac_2O (1 cc.) was added. The clear solution formed was shaken for 10 min. at room temperature. The mixture was poured into ice-water and the precipitate that separated was recrystallized from MeOH-Me_2CO mixture to yield fine pale yellow rods, m.p. 285~285.5° (decomp.). *Anal.* Calcd. for $C_{30}H_{14}O_4(OAc)_8$: C, 60.66; H, 4.17. Found : C, 60.89; H, 4.24. $[\alpha]_D$ −193° (0.2% in Me_2CO).

The authors wish to thank Miss E. Ninomiya and Miss M. Kobayashi for the measurement of infrared spectra, and the member of the microanalytical laboratory of the Institute of Applied Microbiology, University of Tokyo, for microanalyses. One of the authors (K) is grateful to Japan Society for Promotion of Sciences for the grant of a Fellowship. A part of expenses for the present work was defrayed by a Grant-in-Aid for Scientific Research provided by the Ministry of Education, which is gratefully acknowledged.

Summary

The infrared absorption band of C=O in rubroskyrin, which appeared in unexpectedly higher frequency, was discussed in comparison with that of the synthesized model compounds. The presence of alcoholic groups in the molecules of rubroskyrin and luteoskyrin was established by the infrared spectra of O-diacetylrubroskyrin and O-hexaacetylluteoskyrin which showed alcoholic acetate C=O band at 1749 and 1751 cm^{-1}, respectively. These results are incompatible with the proposed structures for rubroskyrin (I) and luteoskyrin (Ⅱ).

(Received February 3, 1960)

[*5] The compound (X) was recovered from the methanolic washing.

[*6] The sample for analysis was dried cautiously at 60~70°, in 2 mm. Hg, over P_2O_5, but partial isomerization into the yellow compound (X) was unavoidable. The analysis was undertaken with the pink leaflets.