Isolation and Identification of Erythroglaucin (1,4,5-Trihydroxy-7-methoxy-2-methylanthraquinone) from the Mycelium of *Alternaria porri* (Ellis) Ciferri†

Rikisaku SUEMITSU, Jyo IWAI,
Kyoko KAWAGUCHI, Noriaki HAITANI
and Noritaka KITAGAWA

Department of Applied Chemistry, Faculty of
Engineering, Doshisha University,
Kamikyo-ku, Kyoto 602

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In order to obtain clues to the biogenesis of hydroxy-anthraquinones, we have investigated on the metabolic pigments of *Alternaria porri* (Ellis) Ciferri, and the isolation of four pigments (I)–(IV) and their mutual relationships on the basis of chemotaxonomy have been reported.¹⁾

In a previous study,²⁾ coloring substances obtained from an ether extract of the mycelium were separated into ethyl acetate soluble and 5% sodium carbonate soluble fractions. These fractions were supposed to contain respectively pigments having at least one hydroxyl group at α -position and those having at β -position in the hydroxyanthraquinones. Generally, only latter pigments in the hydroxyanthraquinones are soluble in sodium carbonate solution. As previously reported,³⁾ three kinds of pigments with *Rf* values of 0.82, 0.45 and 0.37 in TLC have been detected in the ethyl acetate soluble fraction, two of which been identified as 2-methylanthropurpurin-7-methylether (I) and physcion (4,5-dihydroxy-7-methoxy-2-methylanthraquinone) (III) in agreement with the general property

- I: $R_1=R_2=R_3=H$
II: $R_1=R_3=H, R_2=OH$
III: $R_1=R_2=H, R_3=OH$
IV: $R_1=H, R_2=R_3=OH$
V: $R_2=H, R_1=R_3=OH$

FIG. 1.

† Studies of the Metabolic Products of *Alternaria porri* (Ellis) Ciferri. Part VIII. Previous papers are shown in reference 1).

of the hydroxyanthraquinones mentioned above. However, the fraction of *Rf* 0.37 has not been identified yet. This communication deals with the isolation and identification of erythroglaucin (1,4,5-trihydroxy-7-methoxy-2-methylanthraquinone) (V) from the fraction of *Rf* 0.37.

The fraction of *Rf* 0.37 was eluted with methanol to give a crude pigment, which was further purified several times by preparative TLC³⁾ to give deep red plates, mp 202~206°C (lit. 205~206°C,^{4,5)} 202°C⁶⁾) (Yield 3 mg from 306 mg of the ethyl acetate soluble fraction³⁾). In PPC of the pigment, it gave a single spot of *Rf* 0.94 (petroleum benzene saturated with methanol, lit. 0.97⁷⁾), 0.86 (petroleum ether saturated with water, lit. 0.77⁷⁾) and 0.94 (upper layer of acetone-petroleum ether–water, 5:5:3.5, lit. 0.95~0.97⁸⁾). The ethanolic solution of the pigment gave red-violet color by treatment with ethanolic magnesium acetate, suggesting the presence of at least one hydroxyl group at *peri* position to carbonyl group. It gave a molecular ion at *m/e* 300 in the mass spectrum. $C_{16}H_{12}O_6$ (300.26); λ_{max}^{EtOH} nm (ϵ): 229 (5.7×10^4), 255 (2.8×10^4), 275 (2.2×10^4), 302 (sh) (1.2×10^4), 465 (sh) (8.4×10^3), 479 (sh) (9.2×10^3), 490 (1.0×10^4), 510 (sh) (7.7×10^3) and 523 (6.9×10^3); IR ν_{max}^{KBr} cm^{-1} 3100~3600 (broad chelated OH), 1600 cm^{-1} (chelated C=O). In the mass spectrum peaks at 285 (M-CH₃), 284 (M-CH₃-H), 271 (M-CO-H), 270 (M-CH₂O), 257 (M-CO-CH₃), 243 (M-2CO-H), 242 (M-CO-CH₂O), 229 (M-2CO-CH₃), 214 (M-2CO-CH₂O) and 201 (M-3CO-CH₃) were found as characteristic fragment ions of trihydroxy-monomethoxy-monomethylanthraquinone. And three hydroxyl groups are assumed to be located in α positions of the anthraquinone structure, because it had no non-chelated carbonyl band in its IR spectrum.

From the experimental results, this pigment was presumed to be erythroglaucin (V) and the presumption was proved to be correct by the comparison of its UV spectrum with those reported by Matsueda⁶⁾ and Thomson⁸⁾ and its IR spectrum with that of an authentic sample.

In the literature, erythroglaucin was reported as a component of lichen *Xanthoria elegans*⁹⁾ and as a metabolic product of several kinds of *Aspergillus* genus (e.g. *Aspergillus ruber* Thom & Rapper¹⁰⁾). In our experiment, however, erythroglaucin was proved to be a minor metabolic component of *Alternaria porri* (Ellis) Ciferri. As mentioned above, four pigments of (I)~(IV) have been reported to be metabolic pigments of the microorganism. And (V) is a metabolite biogenetically related to physcion (III), and its presence suggests a metabolic pathway from (III) to (V). Moreover, (V) was found to be a metabolic pigment produced by *Alternaria* genus, for the first time. The identification of other minor pigments is in progress.

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