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SHORT COMMUNICATION

BIOSYNTHESIS OF A 9-PHENYLPERINAPHTHENONE BY *LACHNANTHES TINCTORIA*

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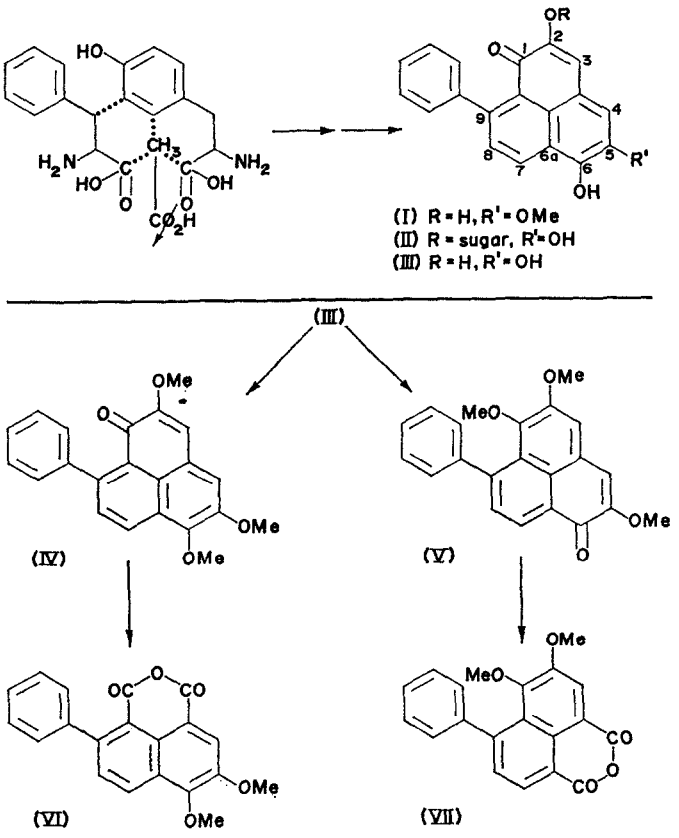
Abstract—The biosynthesis of the 9-phenylperinaphthenone (II) from *Lachnanthes tinctoria*, like that of (I) the aglycone of haemocorin from a related plant, involves condensation of phenylalanine, tyrosine, and acetic acid; the acetate carboxyl carbon atom is eliminated in the process.

9-PHENYLPERINAPHTHENONE pigments, found so far only in plants of the monocotyledonous family Haemodoraceae,[1] differ from the acetate-derived mould perinaphthenones (atrovenetin, herqueinone, etc.)[2] in their unique biosynthesis. Studies by Thomas[3] on the aglycone (I) of the cellobioside haemocorin from Australian *Haemodorum* spp. have confirmed an earlier suggestion by the same author[4] that this compound is formed through combination of one acetate residue with two shikimate-derived C_6–C_3 units. No other example seems to have been described in which one acetate unit participates with two C_6–C_3 units from shikimic acid in the formation of a complex ring system. Both phenylalanine and tyrosine were incorporated; since the label from 2-^{14}C-tyrosine appeared exclusively at C-5, the two amino acids seem to be specific precursors of different parts of the molecule. Scheme 1[3] shows one plausible arrangement of the building blocks, but alternatives in which the acetate carboxyl group furnishes C-6 or C-7 seemed possible.

We have investigated the biosynthesis of lachnanthoside (II), isolated[5] from the roots of *Lachnanthes tinctoria*, the only haemodoraceous plant occurring in the temperate zone of the Northern Hemisphere. Our results (see Table 1) show that here, too, phenylalanine, tyrosine, and acetic acid are incorporated, and that it is the acetate carboxyl which is lost during biosynthesis.

Feeding experiments were carried out on entire young plants, washed free of soil and kept in water to which the labelled precursors were added. Hydrolysis of the glycoside yielded the aglycone (III) (about 3 mg from 10 g of root tissue), which was purified by

[1] J. M. EDWARDS, J. A. CHURCHILL and U. WEISS, *Phytochem.* **9**, 1563 (1970).
[2] A. B. TURNER, *Fortschr. Chem. Org. Naturstoffe* **24**, 288 (1966).
[3] R. THOMAS, *Chem. Commun.* 739 (1971); see also Ref. 2.
[4] R. THOMAS, *Biochem. J.* **78**, 807 (1961).
[5] J. M. EDWARDS and U. WEISS, *Tetrahedron Letters* 4325 (1969); and unpublished work.



SCHEME 1

chromatography on cellulose, recrystallization from chloroform–hexane, and sublimation. Methylation of (III) gave the isomeric trimethyl ethers (IV) and (V), identical with the two dimethyl ethers of (I) described by Cooke;[6] they were purified by TLC (SiO_2; benzene–

TABLE 1.

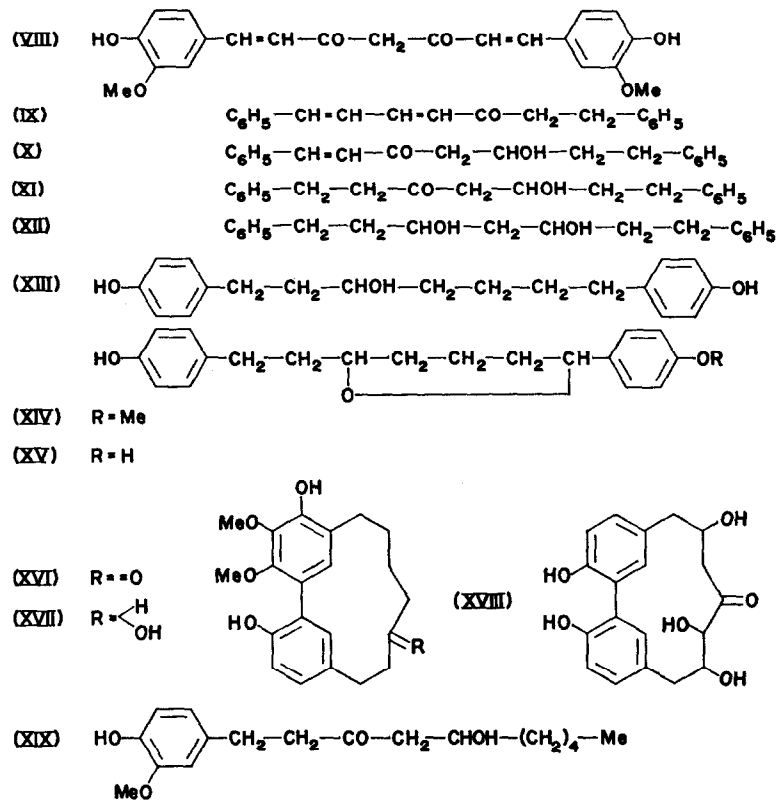
Expt.	Precursor	% incorporation into (III)	Specific activities dpm/μM $\times 10^{-2}$ (III)	(IV)	(V)	(VI)	(VII)
1*	DL-Tyrosine-U-^{14}C	0·062	2·9	2·0	2·1	—	—
2*	DL-Phenylalanine-U-^{14}C	0·449	1·4	1·4	1·4	—	—
3	DL-Tyrosine-3-^{14}C	0·584	2·2	2·5	2·5	2·7	2·2
4	DL-Tyrosine-1-^{14}C	0·465	1·7	1·8	1·6	—	—
5	DL-Phenylalanine-3-^{14}C	3·48	32·5	32·9	35·5	—	—
6	DL-Phenylalanine-1-^{14}C	3·80	23·2	22·9	24·6	—	—
7	Sodium acetate-1-^{14}C	0·236	1·2	1·2	1·4	1·2	1·3
8†	Sodium acetate-1-^{14}C	0·006	—	—	—	—	—
9	Sodium acetate-2-^{14}C	0·62	2·5	2·5	2·5	—	—
10†	Sodium acetate-2-^{14}C	0·082	—	—	—	—	—

A solution of 0·05 mCi of each precursor was fed to the plants.
* Preliminary feeding experiments (1969) on excised roots of older plants.
† 24-hr experiments; all others were run for 3 days.

[6] R. G. COOKE and W. SEGAL, *Austral. J. Chem.* **8**, 107, 413 (1955); **11**, 230 (1958).

phenol, 9:1). In experiments 7 and 9, further degradation of the methyl ethers to the corresponding anhydrides (VI) and (VII)[6] was undertaken, to confirm the specificity of incorporation.

Efficient utilization of phenylalanine, tyrosine, and C-2 of acetate is evident from the data of Table 1. The low incorporation of the acetate carboxyl in the short-term experiment 8 suggests that its relatively high incorporation in the three-day experiment 7 is due to secondary utilization, presumably through reabsorption of expired $^{14}CO_2$. It follows from our results that C-6a must be the atom derived from C-2 of acetate, as shown in Scheme 1. This carbon is hardly accessible to specific degradations; in view of the relatively high incorporations achieved, we hope to investigate this possibility directly with the help of ^{13}C labelling.



It has been pointed out by Thomas[4] that the carbon skeleton of curcumin (VIII), which occurs in *Curcuma* (Zingiberaceae) together with its mono- and bis-desmethoxy analogs,[7] is related to that of (I); although the biosynthesis of (VIII) has not be studied, the close structural analogy strongly suggests a similar origin. Furthermore, recent research has revealed a number of other plant constituents having the same carbon skeleton as (VIII), usually with the aliphatic chain in a more reduced state. Compounds (IX),[8] yashabushi-

[7] R. HEGNAUER, *Chemotaxonomie der Pflanzen*, Vol. 2, p. 465, Birkhäuser Verlag, Basel and Stuttgart (1963).

[8] T. SUGA, Y. ASAKAWA and N. IWATA, *Chem. & Ind.* 766 (1971).

ketol (X),[9,10] dihydroyashabushi-ketol (XI),[10] and (XII)[11] occur in several *Alnus* spp. (Betulaceae), while centrolobol (XIII), centrolobin (XIV), and de-*O*-methylcentrolobin (XV) have been isolated[12] from *Centrolobium* spp. (Leguminosae). The unusual *meta*-bridged biphenyl system found in myricanone (XVI),[13] myricanol (XVII),[13] both from *Myrica nagi* (Myricaceae), and in asadanin (XVIII)[14] from *Ostrya japonica* (Betulaceae) likewise suggests a biosynthesis related to that of I and II. These observations point to the occurrence of the same biosynthetic pathway involving the combination of one acetate with two C_6–C_3 units in quite unrelated plant families; the Haemodoraceae and Zingiberaceae are monocotyledonous, while Betulaceae and Leguminosae are placed in widely separated orders of the dicotyledonous plants. Furthermore, occurrence of compounds such as gingerol (XIX) in the genus *Curcuma* suggests related pathways involving only one phenylpropane unit.

[9] Y. ASAKAWA, F. GENJIDA, S. HAYASHI and T. MATSUURA, *Tetrahedron Letters* 3235 (1969).
[10] Y. ASAKAWA, *Bull. Chem. Soc. Japan* **43**, 575, 2223 (1970).
[11] N. I. UVAROVA, G. I. OSHITOK, A. K. DZIZENKO and G. B. ELYAKOV, *Khim. Prir. Soedin.* **6**, 463 (1970).
[12] I. L. DE ALBUQUERQUE, C. GALEFFI, C. G. CASINOVI and G. B. MARINI-BETTÒLO, *Gazz. Chim. Ital.* **94**, 287 (1964); A. ARAGÃO CRAVEIRO, A. DA COSTA PRADO, O. R. GOTTLIEB and P. C. WELERSON DE ALBUQUERQUE, *Phytochem.* **9**, 1869 (1970).
[13] R. V. M. CAMPBELL, L. CROMBIE, B. TUCK and D. A. WHITING, *Chem. Commun.* 1206 (1970); M. J. BEGLEY and D. A. WHITING, *ibid.* 1207.
[14] M. YASUE, *Nippon Mokuzai Gakkaishi* **11**, 146, 153 (1965).

Key Word Index—Lachnanthes tinctoria; Haemodoraceae; biosynthesis, lachnanthoside; 9-phenyl perinaphthenones.

Note added in proof. After this paper had been submitted for publication, a communication by P. J. ROUGHLEY and D. A. WHITING [*Tetrahedron Letters* 3741 (1971)] on the biosynthesis of (VIII) has become available.